OPTIMAL NET DEPTH FOR OVER-WINTERING BAY D'ESPOIR AQUACULTURE SALMONIDS

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OPTIMAL NET DEPTH FOR OVER-WINTERING BAY D'ESPOIR AQUACULTURE SALMONIDS

by

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ABSTRACT

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An important question for the Newfoundland Salmonid Growers Association (NSGA) is that of necessary net depth for optimal salmonid aquaculture performance efficiency during winter periods when sub-zero water temperatures impose undesirable ongrowing conditions. On the basis of their observations of the past few-years, the NSGA has hypothesized that, given sufficient aquaculture net depth, healthy salmonids will choose a position in the water column that avoids physiologically-difficult conditions. The present document describes the results of a research project undertaken to test this hypothesis and to determine the optimal net depth for over-wintering of Bay d'Espoir aquaculture-industry salmonids.

This research project was designed to examine Atlantic salmon (*Salmo salar*) and steelhead (*Oncorhynchus mykiss*) inventory survival in 10 m vs. 25 m deep nets during winter aquaculture operations. The species factor in this experiment was further divided to include year class (i.e., a typical ongrowing cycle in Bay d'Espoir takes place over an 20-month interval, meaning that at any given time there are two year classes of salmonids in the marine phase of the production cycle). For this experiment, 1^{+}-year fish were from the 2000 year class; 2^{+}-year fish were from the 1999 year class. Odds-ratio tests confirm a significant difference in survival potential between the 10 m and 25 m nets for both salmon and steelhead. However, survival percentages show that this difference is attributable entirely to the 2^{+}-year fish for both species. Odds-ratios indicate salmon in a 10 m deep net are 1.65 times as likely to die during the winter as salmon in a 25 m deep net. For steelhead, the odds-ratio is 1.51. Consequences of over-wintering are much more pronounced among the oldest/largest fish.

Highest mortality was observed among 2^{+}-year steelhead. Percent mortality among these steelhead in the 25 m net to mid-February was 29%. Mortality in the corresponding 10 m net was 45%. Of 20 moribund steelhead subjected to veterinary examination in mid-February, 18 were mature. Many of the moribund fish expelled eggs when handled. This suggests either that the 2^{+}-year steelhead were maturing or that those fish that were maturing were not able to survive the maturation process. Veterinary work confirmed that mortality among the moribund 2^{+}-year steelhead was preceded by osmoregulatory distress. Both cortisol and glucose levels showed the same pattern as blood osmolality.

For the interval of the experiment, Atlantic salmon spent most of their time (i.e., 86% of depth records) at ≤4.0 m in the water column. Steelhead showed somewhat less affinity for these shallower depths but still tended to remain above the 10 m-depth limit of the industry-standard nets. Recommendations to the Bay d'Espoir salmonid aquaculture industry as a result of this research are:

- Use of nets of significantly >10 m for over-wintering of salmonids in high current areas in Bay d'Espoir is not economically justifiable for 1^{+}-year salmonids. Net depths of up to 15 m

may be useful for improving the economic performance of 2nd-year salmonids but only if maturation is controlled and sources of physiological stress can be identified and remedied; and,

- The Bay d'Espoir salmonid aquaculture industry should investigate water-column use by salmonids during warm summer months to determine if nets of 15 m depth have any advantage over 10 m nets during the normal growing season.

Puggon, V. R., A. R. D. Wheeler, T. Nicholls, and D. Whelan. 2003. Optimal net depth for over-wintering Bay d'Espoir aquaculture salmonids. Can Tech Rep Fish Aquat Sci 2000: viii + 55 p.

La profondeur des cages d'élevage de salmonidés permettant d'optimiser la performance des poissons en hiver est une question importante pour la Newfoundland Salmonid Growers Association (NSGA) car les températures de l'eau atteignent un point de congélation créant des conditions peu propices à la croissance. D'après les observations qu'elle a faites au cours des dernières années, la NSGA a formulé l'hypothèse que, si les cages sont d'une profondeur suffisante, les salmonidés en sont chassées une position dans la colonne d'eau leur permettant d'éviter les conditions perturbantes au plan physiologique. Sont décrits dans le présent document les résultats d'un projet de recherche entrepris en vue de vérifier cette hypothèse et de déterminer la profondeur optimale des cages pour l'hivernage de salmonidés d'élevage dans la baie d'Espoir.

Le projet de recherche visait à établir le niveau de survie du saumon atlantique (*Salmo salar*) et du saumon arc-en-ciel (*Oncorhynchus mykiss*) en hiver dans des cages de 10 m par opposition à 25 m de profondeur. La baie espère y en conclure de voir à mener la classe d'âge (c.-à-d. un cycle de croissance typique dans la baie d'Espoir dure 20 mois, ce qui signifie que, à tout moment, deux classes d'âge sont à l'étape de séjour en mer du cycle de production) Aux fins de l'expérience, on a utilisé des poissons d'un an, issus de la classe d'âge 2000 et des poissons de deux ans, issus de la classe d'âge 1999. Un calibrage a confirmé l'existence d'une différence significative dans le potentiel de survie du saumon atlantique et du saumon arc-en-ciel gardés dans des cages de 10 m et de 25 m de profondeur. Les pourcentages de survivants révèlent toutefois que cette différence ne entièrement attribuable aux poissons de deux ans des deux espèces. L'calibrage indique aussi que le saumon atlantique gardé dans des cages de 10 m de profondeur est 1,65 fois plus susceptible de mourir pendant l'hiver que le saumon atlantique gardé dans des cages de 25 m de profondeur. Dans le cas du saumon arc-en-ciel, l'calibrage se chiffre à 1,51. Les conséquences de l'hivernage sont beaucoup plus marquées chez les poissons plus gros et plus âgés.

Le saumon arc-en-ciel de deux ans affichait le taux de mortalité le plus élevé. À la mi-février, celui-ci s'élevait à 29 % dans les cages de 25 m de profondeur, alors qu'il atteignait 65 % dans les cages de 10 m de profondeur. Des 21 saumons arc-en-ciel marbrés soumis à un examen vétérinaire à ce moment-là, 19 étaient morts. Nombre de ceux-ci ont été dans des avec longue manipulée, ce qui laisse supposer que les individus de deux ans étaient en voie de maturation ou que ceux qui étaient en voie de maturation étaient incapables de survivre au processus. Un examen vétérinaire a permis de confirmer que les saumons arc-en-ciel marbrés de deux ans souffraient de détresse osmorégulatrice avant de mourir. Les teneurs en cortisol et en glucose se comparent à l'osmolalité sanguine.

Pendant la durée de l'expérience, la couche algaire a passé la plus grande partie de son temps (> 0,8 W/m² de puissance consommée) à - 0,1 m dans la colonne d'eau. La couche algaire ne se situait donc jamais au niveau ou à une faible profondeur, tout en n'étant pas trop éloignée de la surface pour ne pas être privée de lumière. Une telle profondeur semblait être optimale pour l'activité photosynthétique des algues. Voici les recommandations formulées à l'issue de l'étude effectuée sur la baie d'Elgon :

- L'utilisation de cages beaucoup plus profondes que 10 m pour l'élevage de saumons d'un an dans la partie de la baie d'Elgon où la courant est fort n'est pas justifiée en plus économique. Des cages basses (entre 5 et 10 m de profondeur) offrent pourtant d'excellentes performances économiques des saumons de deux ans, sont esthétiques et le processus de nettoyage est contrôlé et a lieu contre de bons physiologiques pouvant être identifiés et contrecarrés.

- L'industrie salmonicole de la baie d'Elgon devrait établir comment les saumons utilisent la colonne d'eau pendant la nuit dans le but de déterminer si des cages de 10 m de profondeur donneront de meilleurs résultats que des cages de 10 m pendant la saison de croissance estivale.

[illegible] Overfishing is considered one of environmental issues (Healy, n.d., 1993) and its emphasis on ongoing environmental monitoring (i.e., adaptive management), have [illegible] of the most common [illegible] Overfishing industry expansion (Garcia, 1993). Initial emphasis within the early industry on minimizing financial investment, promote [illegible] protection from the [illegible] given way to industry concern about biological and ecological principles [illegible] (Caddy, 1993). The Overfishing industry has progressed well beyond its infancy, having developed considerable weight into its supporting environment, established critical infrastructure to support [illegible] opportunity, and achieved tangible demonstration of its ability to produce quality, inbound products to the international marketplace. Present day Virginia inbound operations [illegible] (2000 tons) [illegible] Atlantic Ocean inbound operations [illegible] (2000 tons) [illegible] inbound operations [illegible] industry, [illegible] operations [illegible] good. Atlantic industry will [illegible] beginning to mature.

In the mid-1980s, the Overfished inbound process [illegible] operations [illegible] Atlantic environment [illegible] Virginia. In response, Overfished Atlantic [illegible] (Healy, n.d., 2000) to six main reasons. First, being located [illegible] Atlantic Ocean [illegible] Virginia [illegible] Second (1985), much [illegible] Overfished [illegible] Third, Atlantic inbound production [illegible] operating 24 months [illegible] months consume, supporting [illegible] inbound operations [illegible] Virginia [illegible] Fourth, Overfished operations [illegible] Atlantic [illegible] Virginia [illegible] Atlantic inbound operations [illegible] Overfished

with an address in the immediate term is that of necessary net depth for livestock survival and optimal salmonid aquaculture performance efficiency during winter periods.

Extensive surveys of winter water thermal characteristics in the Bay d'Espoir area have shown temperatures as low as -1.6 °C. During the winter of 2000/01 the Bay d'Espoir salmonid growers undertook a study to evaluate potential alternative over-wintering sites (NSGA 2001). Thermograph records from North West Cove (East Bay) revealed that winter water temperatures at 7.6 m fell below the lower lethal limit for Atlantic salmon for a brief interval in mid-February. One experimental site, complete with cages of Atlantic salmon, was monitored throughout this interval. In spite of the superchill event, Atlantic salmon survived the winter. This observation led the Newfoundland Salmonid Growers Association (NSGA) to believe that, given sufficient aquaculture net depth, healthy salmonids will choose a position in the water column that avoids physiologically difficult conditions. However, every metre of additional depth for a salmonid cage is an incremental cost factor that must be balanced against improvements in production efficiency. The immediate goal for the salmonid farmers is objective definition of required net depth for effective husbandry. Secondary goals for the project include:

- quantify potential causal mechanisms of fish health influence during winter operations, and,
- elucidate biologically meaningful strategies to refine Bay d'Espoir aquaculture industry husbandry practices under varying environmental conditions.

Ultimately, the industry goal is to develop simple model indicators (response variables) that may be measured at an aquaculture site and applied quantitatively to determine appropriate culture practices (i.e., adaptive management) for application on a day-to-day basis.

METHODS

FARM SITE

The Matchbums (Little Passage, Bay d'Espoir, latitude 47° 37.996'N, longitude -55° 53.591'W; Fig. 1) is an area known in previous years to experience water column temperatures that can be lethal to salmonids during the winter months. This area is one of typically full salinity (i.e., 32 ppt) sea water, though there is an intermittent stream entering the ocean ~1 km from the Matchbums site. This stream occasionally causes some freshwater layering in the upper water column. This site has been licenced for commercial salmonid aquaculture since the summer of 2001.

EXPERIMENT

To facilitate evaluation of cage depth as a means to provide healthy salmonids the opportunity to avoid the extremes of temperature conditions in the water column, a cage array of eight, 7 m x 7 m nets was installed in an area with an overall water depth of ~45 m. This research project was designed around a Mantel – Haenszel odds ratio test (Sokal and Rohlf 1997) to assess Atlantic salmon and steelhead overwinter mortality in 10 m vs. 25 m-deep nets.

during winter operations. The experiment was implemented in the form of a 2^3 complete factorial design (i.e., two species, two net depths and two year classes). The experiment was set up as per Figure 2. As the interest in over-wintering performance is confined within year class relative to depth, comparisons of salmon to trout and one year class to another are of no interest (i.e., differences are of little meaning). Hence, analyses of performance relating to growth and feeding efficiency during the interval are limited to comparisons within species and year class.

Density per cage (i.e., $kg \bullet m^{-3}$) within species and life stage at the beginning of the experiment varied according to the numbers of fish that were available (co-operation agreement with three Bay d'Espoir aquaculture businesses) to apply to the experiment. All densities at the beginning of this over-wintering experiment were kept intentionally low (i.e., $< 9 kg \bullet m^{-3}$) in an attempt to minimize both stress on the fish and potential losses to the farmers. An unexpected compromise to the research endeavour arose when pre-market salmon of the 1999 year class proved not to be available for this experiment. Accordingly, post-spawning Atlantic salmon (grilse) were substituted by the industry participant.

The main performance indicator for this experiment is mortality. Odds-ratio analyses were conducted by species with both year-classes combined. Expected secondary indicators at the time the experiment was implemented were: change in salmon and trout biomass, food conversion (Sveier and Lied 1998) and performance index (McCluskey and Johnson 1958). These response variables were supplemented by blood chemistry measures of physiological status.

MONITORING

Physical

Current was monitored periodically by Acoustic Doppler Current Profiling (ADCP (Workhorse Sentinel, RD Instruments, San Diego). YSI 6600 sondes (hourly recording and archiving of temperature, oxygen and salinity) and Hugrun MS110 thermographs were suspended from the centre of the cage array as per Figure 3 to provide records of the main environmental variables of concern to salmonid farming. The shallowest of the four YSI sondes also recorded pH. CTD profiles (temperature, oxygen, salinity and fluorescence) of the Matchams site, together with an initial ADCP survey of the Little Passage area, were taken on August 30, 2001. An additional CTD profile was taken on April 16, 2002.

The cage array was set up on October 18. Fish were transferred to the site and placed in the respective nets during the interval of November 2 to 15, 2001. The sonde and thermograph array was installed at the site on November 17. This array was retrieved on February 13/02 for uploading of data, re-calibration and servicing. It was re-deployed on February 21. The sonde/thermograph array was removed from the site at the end of the experiment on April 18.

In order to interpret weather conditions of the winter of 2001/02 relative to historical trends for the area, published air temperatures were obtained from Environment Canada for the monitoring station at the Bay d'Espoir hydroelectric facility (47° 59'N, -55° 48'W) for the

interval of 1968 to 1990 (Environment Canada 1994). These data were supplemented with unpublished data, also procured from Environment Canada, for 1991 to 2002 for the same monitoring station. These historical records of air temperature were examined for patterns of winter variability (January through March) relative to water-column temperatures attained from recording thermographs deployed in the Bay d'Espoir area of Newfoundland's south coast during the winters of 2000/01 and 2001/02.

Biological

For this experiment, evaluation of salmonid performance was facilitated by measurements of length and weight at the beginning of the experiment and again at the end. The 35 specimens procured in this manner from each of the experimental groups at the beginning of the experiment were anaesthetized (TMS) then weighed and measured individually and released back to the cage subsequent to recovery from the anaesthetic. Thirty Data Storage Tags (DST; Star-Oddi, Reykjavik, Iceland) were applied on December 5, 2002 to provide hourly measurements of conditions actually experienced by the fish, 20 DST200 series (temperature, salinity, depth) and 10 DST300 series (temperature and depth) tags were applied. Five of the DST200 tags subsequently were reattached to additional specimens. 10 tags were applied to salmon (5 x 200's and 5 x 300's). Twenty tags were applied to steelhead (15 x 200's and 5 x 300's). The model 200 is larger and heavier than the model 300.

All of these tags were affixed to anaesthetized fish with 0.010" diameter stainless steel wire on December 5, 2001 for all but the salmon-grilse cages in order to monitor environmental conditions actually experienced by the fish. Attachment wire was applied through the dorsal musculature by threading the wire through paired (horizontal) 19 gauge hypodermics, inserted through the dorsal body musculature immediately below the origin of the dorsal fin.

Weather permitting, the site was attended every other day through the winter months. Routine activities at the site included observation of fish behaviour, removal and counting of mortalities, recording of secchi disk visibility, hand feeding and record keeping. Salmon and trout in each of the cages were fed to satiation whenever the farm workers visited the site. Satiation was determined subjectively by feeding until the majority of the fish in a cage were no longer actively pursuing the pellets. Underwater cameras were not used at the Matchums site during the winter months. All food-consumption data are based on feed lots weighed both before and after feeding.

Divers visited the site once per week to assure net integrity, record the depth (i.e., direct observation) frequented by most fish in each of the nets and make sure all dead fish were removed and recorded for each of the experimental groups. Dead fish removed from the cages were examined qualitatively for evidence of maturation (i.e., eggs shed during disposal). All mortalities were removed from the site and incinerated at provincially-approved facilities. In addition to observations of maturation at the site, the site technician and NSGA coordinator evaluated the incidence of maturation among the Matchums fish at the time of harvest. These

records were supplemented with processing-plant observations of other Bay d'Espoir farm harvests.

Blood samples were obtained from the 2nd-year steelhead trout on February 18, 2002. The six remaining experimental cages were sampled on April 16-19, 2002. Ten fish from each cage were placed individually in slush ice to simulate actual harvest in an industrial setting. On sedation, blood was collected by cardiac puncture. With the exception of specimens bearing DSTs, all of which were sampled, additional specimens were sampled randomly from each cage. Comprehensive sampling included both tagged and non-tagged fish (8-10 fish/cage) that were anaesthetized using TMS. Fork length and whole weight were measured. Mucus was collected by scraping one lateral side of the anaesthetized fish. Blood was collected in heparinized vacutainers. For the February sampling, blood was centrifuged and plasma subsequently frozen. Mucus was not collected in the February sampling. For the April sampling, blood was analyzed immediately on site using an i-STAT® Portable Clinical Analyzer and CG8+ cartridge (Heska Corporation, 104 Windsor Center Drive, East Windsor, New Jersey 08520). This cartridge measures blood glucose, sodium, potassium, ionized calcium, pH, hematocrit, hemoglobin, total carbon dioxide, bicarbonate, PCO_2, PO_2, O_2% saturation and base excess (BE_{ecf}). After preparing blood smears, samples were centrifuged and plasma collected and kept frozen until analysis.

During the April sampling, both dorsal and lateral skin samples were collected and fixed immediately in 10% formalin and in glutaraldehide (2.5% in 0.1 m sodium cacodylate with 2 mM calcium chloride) for further histological analysis. Liver and muscle samples then were collected and liver weight was recorded. Any gross pathological changes also were recorded.

Plasma total protein concentrations were determined using Sigma total protein reagent (Procedure No. 541). Concentrations of 2, 4, 6, 8 and 10 g•dL^{-1} of Bovine albumin standard (Sigma Diagnostics, Catalog No. P6529) were used as the reference sample. Blood glucose was determined using the Sigma Diagnostics glucose kit (Procedure No. 510). For both total protein and glucose, Beckman Spectrophotometer model No. 25 and Sigma Accutrol Normal and Abnormal controls (Catalog No. A2034 and A3034, respectively) were used. Plasma osmolality was determined using a Fiske One-Ten freeze-point Osmometer. Plasma osmolality was measured for six samples of three pooled specimens each during January, 2002. Plasma cortisol levels were determined by radioimmunoassay (RIA) using Coat-A-Count (I^{125}) Cortisol and CON6 controls (Diagnostics Products Corporation, DPC, Los Angeles, CA). Plasma estradiol concentrations also were measured by RIA using Coat-A-Count (I^{125}) estradiol and CON6 controls (DPC).

Concurrent with the final sampling of salmonids from the experiment, the Matchums site and three additional comparison sites within Bay d'Espoir were examined for presence of harmful microalgae. Niskin bottle water samples (surface, 2 m and 15 m) were taken and preserved in Lugol's iodine solution for subsequent microscopic analysis. Vertical (15 m) plankton-net tows (20μ mesh) were collected and subjected to microscope examination within several hours of procurement. Some of the plankton-tow material also was preserved in Lugol's iodine solution for subsequent analysis.

DATA ANALYSES

Sampling data were subjected to standard distribution analyses (i.e., normality, homogeneity of variance). Environmental data were examined for outliers and any indications that the monitoring equipment might have malfunctioned. DST data were examined for consistency of variable measurements among tags recorded at the same depth at the same time. DST data also were cross referenced with data from the fixed monitoring gear by selecting those observations that fell within ±0.1 m from the depth of each of the fixed thermographs or YSI sondes.

The performance indicators for this aquaculture experiment were intended to focus largely on growth and survival. Data collected during the experiment were examined for overall patterns of performance between cage depths. Comparison statistics were developed on the basis of the following calculations of daily instantaneous rates:

Mortality:

$$Z = \frac{-(\ln N_t - \ln N_0)}{t}$$

where,

- Z is the instantaneous rate of mortality of the group (i.e., the daily change in numbers over the specified time period t);
- N_0 is the number of individuals in the population at the beginning of the specified time interval;
- N_t is the number of individuals in the population at the end of the specified time interval;
- t is the time interval (in this case, days); and,
- ln is the natural logarithm

Growth:

$$G = \frac{(\ln W_t - \ln W_0)}{t}$$

where,

- G is the instantaneous rate of growth (i.e., the mean daily change in specimen weight during the specified time period in days: i.e., t);
- W_t is the mean weight of individuals in the cage at the end of the specified time interval;
- W_0 is the mean weight of individuals in the cage at the beginning of the specified time interval.

t and ln are as above.

With the two quantities of G and Z, the instantaneous (daily) rate of change in biomass was calculated as follows (Ricker 1975):

$$R = G - Z$$

Food Conversion Ratio:
For the present experiment, where appropriate $FCR_{(biological)}$ was calculated as per Sveier and Lied (1998) as:

$$FCR = \frac{FoodTaken_{(Kg)}}{(B_t + B_{dead} - B_0)}$$

where,
B_t is the biomass of fish at the end of the interval;
B_{dead} is the biomass (kg) of fish that died during the interval; and
B_0 is the biomass (kg) of fish at the beginning of the interval.

$FCR_{(economic)}$ was calculated as:

$$FCR = \frac{FoodTaken_{(Kg)}}{\Delta B_{(kg)}}$$

where,
ΔB is the change in biomass between the start and the end of the ongrowing interval.
For both versions of the FCR calculation, whole rather than gutted harvest weight was used.

Growth Coefficient:
Bay d'Espoir industry preference for an alternative way to look at growth of its salmonid strains is included as a form of the thermal growth coefficient (Cho 1992), that often is used by industry as Growth Factor 3 (Holmefjord et al. 1995):

$$GF_3 = \frac{\left(Wt_t^{1/3} - Wt_0^{1/3}\right)}{\sum TU} x1000$$

where,
Wt_0 = mean live weight in grams at the beginning of the time interval
Wt_t = mean live weight in grams at the end of the time interval
ΣTU = accumulated thermal units (i.e., mean daily temperature in °C x number of days) during the time interval.

A challenge in applying these various indicators of strain performance lies in the disparate nature of the data sets from which they are calculated. Accordingly, instantaneous growth coefficients were calculated for the intervals between the sampling dates and then used to calculate mean weight values for each day between sampling dates. This produced a "data" set comparable to the daily inventory and feeding records to facilitate interpretation of FCR and biomass lost to mortality. Water temperature records, consisting of hourly DST measures of

water temperature recorded for the tagged specimens (by species), then were reduced to mean temperature per day and integrated with the derived data set of inventory, mortality, feeding, mean daily weight and FCR to support GF_3 calculations.

The overall performance indicator for this aquaculture experiment is the Performance Index of McCluskey and Johnson (1958):

$$PI = \frac{\Delta Weight_{(kg.)} x100}{FCR_{economic}}$$

where,

$\Delta Weight_{(kg)}$ = increase in total biomass by species and year class during the experimental interval.

In addition to the odds-ratio tests, quantitative analyses performed in the present report focused on weights of the various groups of salmonids from mid-November, 2001 through April, 2002. Pair-wise comparisons were performed using the Student's t-test. All data analyses were conducted with STATISTICA™ V6, StatSoft Inc., 2001.

RESULTS

FARM SITE

From the initial acoustic-doppler survey of the Matchems area, it became apparent that high water currents characterize this tickle (i.e., stretch of water between Long Island and the coastline). Transects across the tickle in the area of the Matchums revealed currents ranging from ~0.5 to 1.7 $m \bullet sec^{-1}$. Subsequent stationary deployments of the ADCP Workhorse in immediate proximity to the Matchums cages indicated currents in the upper 15 m of the water column ranged from 1.0 to 1.8 $m \bullet sec^{-1}$. High current speed was confirmed by observation of billowing of the 25 m deep nets that had to be weighted down with 25 kg. concrete weights, and by diver difficulties in servicing the nets. Water currents at the site were bi-directional, changing in direction each day with the rising and falling of the tide (0.8 to 0.9 m on an approximate 12h cycle; http://www.lau.chs-shc.dfo-mpo.gc.ca/marees/cgi-bin/tide-shc.tcl).

Secchi disk visibility at the Matchums site ranged from 9.0 to 21.5 m. Maximum water clarity was recorded on February 21. Minimum visibility in the water column was recorded on January 21. Throughout much of the interval of the experiment, the depth of most of the fish in the cages was visually readily apparent to site attendants (Fig. 4). Divers at the site reported that the nets, especially the deeper nets, were hanging in the shape of a cone, the lower portion of which (~2 m) was largely collapsed at times of high current. Maximum depth observed by the divers for most of the fish in each cage did not exceed 14 m.

Water and net-tow samples collected in late April at three finfish aquaculture sites in Bay d'Espoir contained the harmful diatoms *Chaetoceros convolutus* and *C. concavicornis*. The

Chaetoceros species were common in the water samples but, at <200 cells•l^{-1}, did not constitute a "bloom" (i.e., > 7000 cells•l^{-1}; Taylor 1993). These harmful species were more abundant at the Roti Bay site but were rare at the Matchems site (Cynthia McKenzie, DFO St. John's, NL, pers. comm.). CTD fluorometer data indicated that maximum chlorophyll on that day was at 2 m.

EXPERIMENT

Although the experiment was laid out as a 2^3 complete factorial design, analyses were limited to the simpler comparisons of start and end weights for each species and year class. To ensure normal data distribution and homogeneity of variance, the weight data were transformed to base 10 logarithms to facilitate quantitative analyses (Table 1).

The experiment lasted for 153 days for all but the 2nd-year (1999 year class) steelhead that were harvested after 92 days. An overview of key over-wintering-experiment results is presented in Tables 2 and 3. It is apparent (Table 2) that there was no significant change in fish size during the winter for 2nd-year salmonids. Shaded cells represent significant differences between start and end weights and show that growth was attained in the 1st-year (2000 year class) fish at all cage depths and in 2nd-year Atlantic salmon only in the 25 m cage. Mean-weight characteristics between the sampling periods for 1st-year fish are illustrated in Figure 5.

MONITORING

Physical

CTD profiles of the Matchums site, together with an initial ADCP survey of the Little Passage area, plus the fixed recording thermographs and YSI sondes, served to characterise biologically-meaningful variables of water column characteristics through much of the period of the experiment. The YSI unit at 19 m apparently became unstable toward the end of January. With the exception of this unit for February onwards, the fixed monitoring gear provided good data on the state of the marine environment at the Matchums site. The CTD profile of Figure 6 (April 16) is representative of salinity and temperature stratification at the Matchums. The depth of the lower-salinity surface waters varied considerably with rainfall but typically extended to somewhat <5 m in the water column. Examination of water column characteristics at 3 m depth (Fig. 7) showed considerable fluctuation leading up to the February termination date for the 2nd-year steelhead. Salinity variation at this depth was confined to the range of 26 to 34‰.

Water temperature data consisted of hourly measures of water temperature at the depths of the fixed monitoring gear (nine units, Fig. 3). These data were reduced to mean temperature per day and plotted by depth for comparison with the DST depth data. For the upper 10 m of the water column, temperatures from the fixed gear throughout the experiment ranged from a minimum of -0.65 to maximum of 8°C. Salinity at 3 m varied from 26.3 to 34.1‰, dissolved oxygen from 8.4 to 14.1 ppm and pH (3 m only) from 7.8 to 8.0. Water temperature reached its lowest level in February. During February, oxygen at 3 m varied from 10.5 to 11.5 ppm,

salinity from 29.4 to 32.0‰ and pH from 7.82 to 7.97. The fixed monitoring array confirmed occasional negative temperatures in the upper water column (i.e., ≤ 6 m) in February and March but did not show evidence of superchill (i.e., <-0.7°C). Water temperature at 1 m dipped below zero for the first time on February 5. Interpretation of thermal units from the nine fixed monitoring units for the October/01 through April/02 interval is presented in Appendix 1.

For comparative purposes, to interpret the water column temperatures at the Matchums relative to other sites in the Bay d'Espoir area, thermograph records collected by the provincial Department of Fisheries and Aquaculture during the winters of 2000/01 and 2001/02 were examined for frequency of sub-zero temperatures. Of the eight locations identified in Table 4, all experienced superchill conditions at some level in the water column.

Having established the conditions that prevailed at the Matchums site during the winter of 2001/02, more specifically the seasonal minima that occurred, and the thermal characteristics of other locations in the Bay d'Espoir area, the question remained as to how such winter conditions compare with historical trends in climate. Figure 8 presents historical air temperatures for the coldest winter months (January through March) for the area for the interval of 1968 to the spring of 2001.

Biological

During the interval of the experiment the fish received an average of about 20 kg. of feed every second day for a total of ~13.5 t of food overall for the winter period (Table 5). Considering for the most part that 2nd-year salmonids during the interval of the experiment did not increase in size, daily instantaneous growth calculations (Table 6) become moot for this year class. In contrast, significant differences in live weight of 1st-year salmonids between the start and end of the experiment (Table 2) suggest further consideration of performance indicators is in order for these 1st-year fish.

Of the 30 specimens fitted with DSTs on December 5/01, one escaped from the anaesthetic recovery bath. Of the 29 tagged specimens remaining, 21 were recovered at the end of the experiment. One of the tags was completely flooded. During the course of the experiment, divers in the cages recovered five shed tags. For these tags, it was apparent that the stainless steel wire had snapped at the point of attachment to the fish. These recovered tags were reapplied to other fish. Of the 21 tags recovered at the end of the experiment, seemingly meaningful data were obtained from 19 fish. In all, there were 25 DST200 and 10 DST300 deployments (including redeployments). Recovered tags included all 10 DST300's but only 12 DST200's (including a flooded tag). Tag loss was much higher in the 200 series than for the smaller 300 series tags. Over all, the recovered DSTs provided 37339 data records. This data set is composed of 21531 records of temperature, depth and salinity (i.e., the 200 series tags). About 57% of the recovered records are from 200 series tags though ~70% of the tagging was done with the 200 series.

The DSTs applied to Atlantic salmon resulted in ~28000 observations of the variables of interest (mean of 2875 records per variable per tag). For the trout, the DSTs provided ~14000 useful records per variable (mean of ~1200 records per variable per tag).

On initial scrutiny of the DST records, it became obvious that, for some DSTs, depth data "correction" was in order. For those tag units in which depth records suggested flying fish, all depth data were adjusted so that minimum depth was zero (i.e., surface). After depth adjustment, DST data records from 19 fish provide an indication of the water column frequented by the tagged specimens (Table 7, Fig. 9). The DST data set then was examined by depth and time to identify those measurements recorded at the depths of the fixed monitoring gear whenever these specific depths were recorded among different fish at the same time. This process revealed 1198 records (i.e., minimum of two records, maximum of five records observed at the same depth at the same time, all within the upper 6 m of the water column), on which to base an evaluation of the consistency of measurements among tags. Temperature correspondence among tags typically was within 0.5°C (Table 8). Maximum discrepancy recorded was 2.0°C. Salinity proved somewhat more variable among tags recorded at the same time at the depth of the environmental monitors. Graphs of maximum and minimum observations (same time, same depth) for salinity and temperature (Fig. 10) suggest at least one of the salinity measures towards the end of the experiment is questionable.

On confirmation of compatibility of measured variables among tags, the DST data were compared with those from the fixed monitoring gear. Graphs of DST data, superimposed on the depths in the water in which there was fixed-monitoring gear, suggest some discrepancy, more so for the salinity data. However, the general overlap of the data (Fig. 11), suggest that the variables recorded by the DSTs were of approximately the same magnitude and followed the same time progression as that of the fixed monitoring gear. As a supplement to the environmental data for the water column provided by the fixed monitoring gear, we interpret the DST records provide further insight into conditions experienced by the fish (Table 9). For the interval of the experiment, the minimum temperature recorded via DST for salmon was -0.8°C, at which time (February 13) the specimen was at 2.0 m. For steelhead, the minimum temperature recorded was -0.9°C (also on February 13), at a depth of 1.5 m.

Further examination of DST data suggests a perspective more strategic to the goal of this research project. Figure 12 illustrates depth distribution of the Matchumm salmonids relative to water temperature for the months of January through March. For Atlantic salmon, 86% of the depth records were from ≤4.0 m, 13% between 4.0 and 15 m and <1% from >15 m. For steelhead, 60% of the depth records were from ≤4.0 m, 38% between 4.0 and 15 m and 2% from >15 m.

For all of the fish fitted with DSTs, tissue damage at the site of the tagging wound proved extensive. This appeared more pronounced among the trout than the salmon. However, at least for the duration of this experiment, the tagging wounds did not result in mortality. Four of the 29 tagged specimens in the cages died during the term of the experiment. Specimens with the DSTs typically were observed milling with the rest of fish and did not show any obvious behaviour patterns that differed from the untagged fish. Fixed monitoring gear provided data on

temperature at depths to the bottom of the deepest nets. Recognizing that salmonid depth distribution most often ranged through the upper 10 m of the net column, Figure 13 depicts temperature fluctuations only for depths ≤10 m. What is apparent from the daily temperature ranges is that temperature fluctuation diminishes with depth.

Percent mortality among the 2nd-year steelhead in the 25 m cage to mid-February was 29%. Mortality in the corresponding 10 m cage was 45%. Moribund fish typically ended up towards the surface of the cage, seemingly disoriented and swimming on their sides. Approximately 90% of the moribund fish expelled eggs during veterinary examinations.

Of 20 moribund steelhead subjected to veterinary examination in mid-February, 19 were mature. Plasma osmolality of pooled samples was as follows: 504, 615, 634, 650, 604 and 615 mosM•kg^{-1}. Due to the extreme financial implications of the escalating mortality in this group, the owner harvested all the 2nd-year steelhead on February 19/02 (i.e., total of 92 days for this year-class component). Observations of the harvested steelhead at the processing plant confirmed eggs in ~10% of the harvest. Of the 2nd-year steelhead harvested for the overall industry from the pre-market holding area at St. Alban's, once again ~ 10% were identified either as mature or maturing.

Other Performance Measures

Calculations of monthly mean $FCR_{\text{accum. nitrogen}}$, accumulated thermal units and GF_3 are represented in Appendix 2. Even with the evidence of growth for 1st-year fish, it is apparent that the biomass of fish lost to mortality occasionally outpaced gains in biomass attributable to growth. In such situations, calculated values of $FCR_{\text{accum.}}$ have little meaning and therefore have not been included in the appendix. $FCR_{\text{accum. gain}}$ calculations for steelhead showed a generally-improving trend through the duration of the experiment (Fig. 14).

Blood osmolality, cortisol and glucose levels were significantly higher ($p = 0.0001$, 0.00178 & 0.00222 respectively) in 2nd-year steelhead trout samples obtained in February than all trout and salmon samples obtained in April (Fig. 15). Appendix 3 contains physical measurements and blood-analysis results of the comprehensive sampling. Only physiologically and/or statistically-significant results are presented.

No results were obtained for estradiol for Atlantic salmon samples. Circulating levels in the blood may have been too low for the analytical procedure applied. For steelhead trout, two of the 2nd-year specimens were found to contain mature eggs during sampling. High estradiol concentrations were apparent (8078.92 and 6508.28 pg•ml^{-1}, 10 m and 25 m deep cages, respectively). These extreme high values were excluded from the analysis. Table 10 shows that the 1999 year-class samples obtained in February had significantly lower estradiol levels than the 2000 year-class samples obtained in April ($p = 0.0002$).

[illegible]

[illegible]

Salinity fluctuations at 3 m depth (i.e., 20 to 30‰, Fig. 7) span a range from brackish to full salinity marine conditions ([illegible]). The DST subset from the levels of the fixed monitoring gear (Fig. 12) and CTD (Fig. 6) salinity records however demonstrate that salinity above 3 m was subject to freshwater layering, most likely from the river by waterfall. For Atlantic salmon, overall there were two DST salinity records of <10‰. For steelhead, there were eight such records. By far the greatest frequency of salinity records for both species fell in the range of 20 to 30‰.

Many of the moribund fish spent much of their time close to the surface than is normal for a healthy salmonid stock. This is a common behaviour for sick and dying fish. Coupled with the veterinary observation that many of the moribund fish expelled eggs when handled, the observations suggest either that the 2nd year steelhead were maturing or that those fish that were maturing were not able to survive the maturation process, possibly due to osmoregulatory failure. The elevated serum osmolalities (i.e., >600 mmol/kg^{-1}) determined for the February samples of moribund steelhead support this but argue against the presence of a significantly lower salinity layer at the Wiachuma site. Isotonic salinity is variously reported in the literature for salmonids as 9 – 10‰ or ¼ sea water (Morgan and Iwama (1990), D'amaschi et al. (1993), Morgan and Iwama (1991)). Such low salinity conditions, at which osmoregulatory demand is minimal, were not recorded at the Wiachuma site. However, the salinity variability of the upper water column may have been sufficient enticement to draw maturing fish toward the surface in search of osmoregulatory relief, only to subject them to occasionally lethal water temperatures, the greater temperature variability characteristic of the upper water column of the aquaculture nets (Fig. 17), and the greater likelihood of encountering ice crystals.

Thermograph records for the Wiachuma indicate a +30 degree day advantage in thermal units in 11 to 17 m water relative to the thermal characteristics of the 4 to 11 m range (Appendix 1). It is apparent from the temperature ranges recorded per day through the experiment (Fig. 17) that there is lower diurnal temperature fluctuation with increasing depth as well as elevated minimum water temperature. What appears as both a slight thermal growth advantage and reduced diurnal temperature variability to additional net depth does not appear to have been utilized very often by the fish (2% of depth observations for salmon, 15% for steelhead).

[illegible]

The odds ratio tests (both year classes combined, Table 5) indicate a significant difference in survival potential between the 11 m and 25 m nets for both salmon and steelhead. However, survival percentages confirm that this difference is attributable to the 2nd year fish (i.e., 1999 year class) for both species. Odds ratios indicate that salmon in a 11 m deep net are

[illegible] cases in likely to be during the winter as shown in [illegible] in deep sea. For seafood the consumption [illegible] Again, when examined in the context of the present survival among the groups, it is apparent that survival advantage of increased net depth is achieved only among the [illegible] year fish. It is apparent that environmental influences as ever evening survival are much more pronounced among the older/larger fish. Thus, for [illegible] year fish of both species survival is enhanced significantly by the use of deeper nets.

MONITORING

Physical

In light of the data of Table 8, it is apparent that marine especially is an occasional water phenomena of the Bay d'Espoir area, though typically of short duration and most often at depths ranging to 4 m. With the exception of the Hatchet Cove observation, such conditions are infrequent. Given the observations of the present report and those of NSOUR (2001), winter especially events were not associated with any obvious elevated mortality. It is apparent that aquaculture salmonids can survive some especially events. However, this observation should not be a basis for overconfidence. The presence of ice crystals in the upper water column, coincident with turbulence, the absence of [illegible] ice cover and especially conditions in the upper 0 m of the water column, represents a combination of variables likely to be lethal to aquaculture salmonids. Such conditions have been encountered even in the relatively protected area of Bon Bay and resulted in extreme mortality among the rainbow trout at the site (C. Collins, Wetland Aquaculture, pers. comm).

The Hatchet Cove water column temperature characteristics of the winter of 2002, though somewhat warmer overall, are not inconsistent with those of other cold coast marine areas during the winter of 2001 (Table 8). Two concerns arise if the winter warming trend of the mid 1990s (Fig. 8) were to continue towards re-establishing winter norms of the previous two decades. First, increasing winter temperatures likely would encourage increased ice melt in northern latitudes, potentially increasing the flow of the Labrador current. Such increased flow would serve to decrease marine temperatures of coastal Newfoundland during the summer and could result in increased ice transport to south coast marine areas. This has implications to salmonid aquaculture.

Biological

Feeding data (summarized in Table 7) suggest that on average, there was little difference in food consumed per individual salmon relative to cost. However, these data need to be considered with the caveat that the over-wintering operations at the Hatchet Cove combined without the benefit of underwater cameras to confirm the incidence of uneconsumed pellets reaching the bottom of the nets. Hence the feeding data may not be totally reflective of consumption. Taken at face value, the data suggest on average, salmonids in 10 m deep cages consumed more food per individual than salmonids in the 25 m deep nets (i.e., > [illegible] vs

...(depth)...

...

...

might challenge salmonid swimming ability. In consequence, over-wintering salmonid distribution patterns may have been influenced more by net movement than by current speed.

Aside from the obvious problems experienced with the stainless steel wire used to attach the DSTs to the fish, the results obtained have been instrumental in documenting winter water column use by these aquaculture salmonids. Correspondence of DST data among tags (Fig. 10) and from side overlap with data of the fixed monitoring gear (Fig. 11) suggest the DST data are meaningful. In future, should it prove useful to use this DST technology again, a heavier gauge of wire would be appropriate (i.e., MONEL® 0.020" nickel/copper). Overall the 200 series tags provided good results when they worked, but were prone to failure. In future, except at locations in which salinity is expected to fluctuate greatly throughout the water column of an aquaculture cage, use of the smaller 300 series DSTs may be more appropriate and may prove less damaging to the fish.

It appears that a 25 m deep net is unnecessary for the Matchbox marine environment and in fact, not well suited to this high-current area. Maximum depth recorded for specimens that bore the DSTs in 25 m nets was 23.2 m. However, DST-recorded depths (Fig. 12) frequented most often by the fish during the coldest months of the winter were <4 m. (86% for salmon, 60% for trout). Though there may be a survival advantage to deeper nets among 2+-year steelhead (Table 3), the DST data suggest that these aquacultured salmonids rarely occupied depths >15 m. This is consistent with observations of the cage attendants and the divers (Fig. 4). Given access to deeper nets, the fish occasionally (i.e., <6% of DST observations overall) ventured below the depth limits of the 10 m nets. As implied in Fig. 13, diurnal temperature variability decreased with increasing depth. However, the pattern of fish depth distribution relative to water-column temperature for the DST data (Fig. 12) suggests that low winter water temperatures did not encourage these aquacultured salmonids to seek greater water depth. Alternatively, it may have been that reduction in the cross-sectional area of the nets at greater depths imposed a constraint to salmonid depth range that otherwise may have been expanded by low temperature. When considered relative to the superchill records of Table 4, it appears the "at-risk" levels of the water column typically are within the upper 8 m. This being the case, were an entire net inventory to seek greater depth during unfavourable thermal conditions, a 10 m.-deep net would impose a water-volume constraint. Under such conditions, a 15 m.-deep net may well provide a safety zone that, even if rarely used, would constitute a refugium of considerable survival value to the fish.

From both the diver observations and the DST data, it is apparent that calculations of fish density, based on overall cage volume, represent minimum possible figures for kg•m^{-3}. On the basis of the observations of Figures 4, 9 and 12, depth preference (i.e., most of the data most of the time) was similar for 1+-year fish irrespective of species, typically falling in the range of 1.1 to 4.9 m. The only data for 2+-year fish (i.e., steelhead, 25 m net) indicated a depth preference of 4.2 to 6.9 m.

The mitigating effect of net depth on mortality of 2+-year steelhead to February (i.e., 67% mortality for 10 m net vs. 29% for 25 m net), and the absence of such an effect for 1+-year salmonids, leads to questions of the physiological demands of over-wintering conditions and the

mechanism by which mortality might be mediated. Water and plankton net-tow samples collected in late April at finfish aquaculture sites in Bay d'Espoir contained the harmful diatoms *C. convolutus* and *C. concavicornis*. Both of these species have barbed spines that can irritate the gills of penned finfish. This irritation and resulting mucus formation has led to high mortality rates of cultured finfish in British Columbia (Rensel 1995). While *Chaetoceros* species were common in the April water samples at the Matchums, the number of cells per unit volume did not constitute a "bloom". We suspect, during the cold-water interval of February, that the incidence of *Chaetoceros* in the water column would have been even less than in April and therefore unlikely to have contributed significantly to the observed mortality. At the time of the April plankton tows, these harmful species were found in greater abundance in Roti Bay, a favoured industry over-wintering site. They were at a low density at the Matchem site. The presence of these diatoms in Newfoundland waters near finfish aquaculture sites confirms the potential for *Chaetoceros* blooms here that are similar to those that have been found in British Columbia. With continuing development of the finfish aquaculture industry in Newfoundland, it becomes increasingly important to assess the risk posed by these harmful-phytoplankton species and determine how improved site selection and management may be effected.

For five of the eight cages in the experimental design, the biomass of salmonids remaining at the end of the experiment was less than the starting biomass (Table 2). By discounting the 2nd-year salmon (i.e., grilse), the diminishing biomass overview reduces to three of six cages. Biomass loss in 50% of the experimental groups represents poor over-wintering performance. For those months in which FCR_____ calculations were derived from meaningful data, the values ranged from reasonable to unacceptable (Fig. 14). Comparison of the economic and biological FCRs (Appendix 2) confirms that mortality was the predominant factor in this undesirable situation. In contrast to the 2nd-year salmon that already had matured and therefore were expected to incur elevated mortality, loss of 2nd-year steelhead was unexpected.

A poor second to the challenge of improving over-wintering survival is the question of poor (or no) growth during the winter months. The analysis of only modest growth for the 1st-year salmonids renders consideration of food conversion ratios and performance index (Appendix 2) a questionable exercise. It is apparent that a maintenance ration is necessary to sustain fish through the winter and that there is a significant cost to this aspect of Newfoundland salmonid farming. An issue that becomes significant is that of the benefits of carrying salmonid inventory through a second marine winter relative to harvesting at less than two years of marine engrowing. Given the physiological consequences of maturation and the level of mortality documented among moribund fish in the present study, it is apparent the potential benefits of larger fish to the marketplace were negated by lost production. While compensatory growth (Nicieza and Metcalfe 1997, Reimers et al. 1993) during the subsequent growing season might offset these losses, it is the excessive mortality problem that must be remedied. Net depth alone is not a solution to this problem.

STRESS AND OSMOLALITY DISTURBANCES

High mortality reported for the 1999 year-class steelhead in February 2002 was attributed to osmolality disturbances. While April samples showed normal osmolality figures (means of 346.9-361.0 mmol•kg^{-1}, Appendix 3) for both steelhead and salmon (normal range in rainbow trout is 296-340 mmol•kg^{-1}, Bowser 1993), the February samples showed pronounced osmotic disturbances. Both cortisol and glucose levels showed the same pattern as osmolality, with maximum concentrations up to 37.8 µg•dL^{-1} cortisol and 378.0 mg•dL^{-1} blood glucose. This indicates that the 2nd-year steelhead were under stress in February (normal blood glucose in rainbow is 63-144 mg•dL^{-1}, Bowser 1993).

Apparently, osmolality disturbance of the 2nd-year steelhead was a reflection of that stressed state. Stress in fish results in two types of endocrine response, the adrenergic response, resulting in increased plasma concentrations of adrenaline and noradrenaline, and the hypothalamo-pituitary-interrenal (HPI) response, culminating in increased plasma cortisol concentrations (Sumpter 1997). Basal cortisol concentrations in unstressed fish generally are reported to be between <1 and 10 ng•ml^{-1} and rise very rapidly in response to stress. Generally, the first detectable rise occurs around 5-10 min after initiation of stress and concentrations continue to rise, as long as the stress is sustained, to reach between 100 and 200 ng•ml^{-1} after about 1h, (Sumpter 1997).

Secretion of cortisol influences both mineral and carbohydrate metabolism in fish (Donaldson 1981). Both rainbow trout and coho salmon subjected to handling stress show hyperglycemia and hyperchloridemia with peak values being reached after 3-4 h, returning to basal levels after 24h (Wedemeyer 1972). Blood glucose increases as the stress response begins in order to provide energy to the animal for the 'fight-or-flight' reaction. Elevated blood glucose levels are initiated and sustained by the actions of adrenaline and cortisol, respectively, on the liver and muscle (Morgan and Iwama 1997).

Physical disturbances of rainbow trout adapted to sea water or dilute sea water increase both the plasma sodium ion (Na^+) concentration and the Na^+ efflux and influx. Lahlou et al. (1975) have shown that under these conditions the Na^+ efflux may double to ~10 mM•kg^{-1}•h^{-1} and the Na^+ influx may increase 7-fold to 20 mM•kg^{-1}•h^{-1} (Eddy 1981). Given the necessity for site maintenance (i.e., feeding, removal of mortalities, net inspection and repair), plus excessive current at the site that resulted in obvious net billowing, it is apparent that some physical disturbance of livestock is unavoidable. However, the most obvious candidate for mitigation is a lower-current site where net billowing is minimized.

The question that needs further study is what causes the stress for the 2nd-year steelhead back in February? Is early maturation to blame? Unfortunately, samples were collected from the affected groups only in February. Such one-time sampling is insufficient to resolve this question.

Early Maturation

Veterinary work confirmed that mortality among the moribund 2nd-year steelhead was preceded by osmoregulatory distress. Blood plasma osmolality of marine-adapted salmonids typically falls in the range of 300 - 400 mosm•l^{-1} (Byrne et al. 1972, Staurnes et al. 1990, Rørvik et al. 2000). Samples from the Matchums 2nd-year steelhead, with serum osmolalities >600 mosm•l^{-1}, indicate that steelhead muscle tissue was severely dehydrated. Similar observations have been made elsewhere (Allee 1981). It appears that the osmoregulatory mechanism had failed and the salinity of water at the site was drawing the body fluids out of the fish. Observations during veterinary monitoring suggested maturation rates among moribund fish of >90%. Observations of the Matchums steelhead at the processing plant confirmed eggs in ~10% of the harvest. Of the 2nd-year steelhead harvested for the overall industry from the pre-market holding area at St. Alban's, only ~10% were considered to be either mature or maturing.

When viewed in the context that eggs were observed in only ~10% of the harvested fish at the processing plant, the observation of >90% maturation among moribund fish at the Matchums suggests that mortality was attributable largely to factors associated with reproductive development. The lower incidence of maturation among the 2nd-year steelhead observed at the fish plant may be due to excessive mortality among maturing specimens prior to the time of harvest. The apparent discrepancy between these observations suggests that the maturing fish were dying and that the fish left at the time of harvest were those less advanced in their reproductive cycle.

Of equal concern to mortality among maturing 2nd-year steelhead is the observation of a low level of maturation among 1st-year steelhead. Early-maturing salmonids, occasionally even 1st-year fish, were found during sampling in April, 2002 and have been reported by the industry and provincial aquaculture veterinarians in previous years. Sexual maturation of farmed salmon before harvest in seawater is undesirable to farmers, since it is associated with a cessation in growth and decline in fish quality, which reduces market value (Stead et al. 1999). Based on hormone analyses, it is apparent that salmonid production efficiency is compromised by advancing reproductive development.

In fish, as in other vertebrates, sexual development is endocrine mediated via the hypothalamic-pituitary-gonadal axis. Development of the gonads is under control of hormones from the pituitary, that in turn is controlled by the hypothalamus and by hormone feedback, especially by gonadal steroids (Stead et al. 1999). 17β-estradiol (E2) is the principal ovarian steroid in female salmon that plays a physiological role in gonadal differentiation, growth, gametogenesis and maintenance of somatic tissue (Fostier et al. 1983). In female trout, low levels of E2 have been measured during the period of slow oocyte growth. Maximum values for E2 have been determined during vitellogenesis (Scott et al. 1980, Schulz 1984).

17β-estradiol is the only hormone measured in this study. Estradiol level in Atlantic salmon samples was undetectable with the method used. Either the fish had a very low level of the hormone or the method was not suitable to be used with the salmon samples. The Coat-A-Count Estradiol kit has been used previously in rainbow trout (Pereira et al. 1998). The assay

has been validated for *Oncorhynchus mykiss* by comparison of estradiol either in plasma or in estradiol fractions using column chromatography on a Sephadex LH-20. Pereira et al. (1998) found that the antibody for estradiol was highly specific, showing a relatively low cross-reactivity to other natural-occurring plasma steroids.

The causes of early maturation need further study, as evidenced by the fact that even selected Norwegian-origin strains in British Columbia are showing increased incidence of early maturity (Brian Glebe, DFO, Saint Andrews Biological Station, pers. comm.). Stead et al. (1999) provide evidence that growth and sexual maturity in Atlantic salmon are closely linked processes. This makes it difficult to determine if accelerated growth induces maturity. The body fat reserves and/or lipid content in the diet also may influence early maturation, especially in males. Rowe and Thorpe (1991) showed that in male Atlantic salmon parr, maturation is suppressed when mesenteric fat fails to exceed an undefined level by May. Other studies showed that the percentage of maturing male Chinook salmon was significantly influenced by whole-body lipid, increasing from 34% in fish fed 4% lipid diet to 45% in fish fed 22% lipid diet (Shearer and Swanson 2000). Furthermore, the dietary polyunsaturated fatty acids (PUFAs) may have an effect on maturation. Asturiano et al. (1999) suggested that PUFAs and their derived PGs modulate oocyte maturation in the European sea bass. In the testis, they appear to regulate the synthesis of the prostaglandin E2 (PGE2 - a general modulator of "local" hormones), androgens and progestagens.

The Bay d'Espoir industry practice of towing cages of pre-market fish to the processing plant further up the estuary, and subsequently culling maturing fish from the freshwater lens by seining (Pepper et al. 2001), confirms the preference of maturing salmonids for lower-salinity water. These observations suggest that the maturing steelhead were physiologically compromised in their ability to deal with high salinity, an interpretation that is corroborated in the literature (Ulgenes and Naevdal, 1986). The combined influence of dirunally-fluctuating, low water temperature in the top two metres of the water column (Fig. 13), together with reproductive development encouraging the fish into water with lower osmotic demand, thus may be a source of mortality among the 2nd-year steelhead.

In view of the observations of this over-wintering research, the Newfoundland industry is left with two options: 1. Determine the extent of livestock maturation and address the maturation issue; or, 2. avoid over-wintering 2nd-year salmonids. Considering that some maturation has been documented even among 1st-year steelhead, it would seem prudent that industry adopt the former tactic. Strategies to deter maturation need to be developed. As noted by Isaksson (1991), there have been pronounced increases in early maturity following mild-winter temperatures, even in selectively-bred Norwegian stocks. While manipulation of water temperature is not likely feasible for marine-cage aquaculture operations, manipulation of photoperiod, diet and ration level to deter maturation (Ingram 1985) may be an option.

As recorded by Pepper et al. (2001), the amount of Atlantic salmon production lost to the Bay d'Espoir industry due to early maturation has been increasing during recent years. Reasons for this increase are unknown but are expected to be a consequence of both breeding and husbandry practices and of environmental conditions during the milder winters of the late 1990s.

As becomes apparent from Figure 8, Bay d'Espoir winters were relatively warm through the 1970s and '80s, hit a severe low in 1991 and, generally, have been warming up since then. It appears that winter conditions of more recent years have been closer to the norm than to the extreme. Continuation of this trend has implications to Bay d'Espoir salmonid aquaculture. The husbandry context of generally-mild winters, and physiological conditions for early maturation are described by Saunders et al. (1983). These observations highlight the need for a better understanding of the physiological basis for winter-husbandry practices and further work with broodstock management.

CONCLUSIONS

In some ways, the results of the present experiment are not those expected by the salmonid farmers. The assumption was that aquaculture salmonids would go deeper in the water column to avoid harsh winter conditions. Figure 12 suggests this was not the case and that, irrespective of temperature, salmonids tended to remain in the 2 m to 4 m wedge of the water column. The deeper nets do not appear to have conveyed any winter advantage relative to avoidance of sub-zero temperatures, though there was lower mortality among 2nd-year salmonids in the deeper nets. What still remains questionable is whether deeper nets convey any advantage to summer farm operations and if deeper nets are a liability under summer conditions of significant current.

It has been a common observation among Bay d'Espoir salmonid farmers that in summer the fish, except when feeding, go deeper in the water column. Such behaviour has been documented elsewhere (Fernoe et al. 1995). Whether or not this is avoidance of elevated upper water-column temperature (or light intensity or lower salinity) is not known. However, given that the growing season is the time when performance has to be optimal in order to meet the financial prerequisites of an aquaculture business, evaluation of optimal net depth under summer conditions seems warranted. Temperature in the upper water column of Bay d'Espoir during August can reach 20°C. Dwyer and Piper (1987) recommended growing conditions for Atlantic salmon be regulated to the range of 13°C to not more than 16°C. As a result of the present experiment, much of the necessary infrastructure is in place to support evaluation of optimal-net depth during the growing season. Unlike the suboptimal environmental conditions that were expected to prevail at the Matchums during this over-wintering experiment, there are not likely to be significant risks to conducting similar research on optimal-net depth during the summer months. Given that much of the asset infrastructure already exists with which to undertake such work at a summer ongrowing site, it could be beneficial to future industry economic viability to consider further research on optimal-net depth for Bay d'Espoir aquaculture. In consideration of the Matchums experience with its obvious confounding variable of current speed, were such a study to be pursued by the Newfoundland salmonid growers, such efforts should focus on a lower-current site.

Based on the data of this report, it does not appear economically justifiable to adopt aquaculture nets in Bay d'Espoir that are significantly greater than 10 m deep for over-wintering salmonids, especially in high current areas. This does not preclude potential advantages to

additional net depth for summer operations. At least during the winter months, it is apparent there is no advantage to net depths >15 m. DST technology was instrumental in documenting salmonid depth preferences in the over-wintering nets. This technology may be appropriate also for evaluating depth preferences during the summer months.

What appears critical to further industry development is a program to profile salmonid gonad development towards identifying husbandry practices oriented towards deterring maturation. As noted in the introduction to this report, there is a significant industry need for simple indicators that may be measured at an aquaculture site and applied quantitatively to determine appropriate culture practices for application on a day-to-day basis. This goal has not been achieved in the present study. However, *in situ* measurement of gonad size, plus blood measures of osmolality, cortisol, glucose and estradiol are obtainable and may prove useful in developing approaches to salmonid maturity control for the Bay d'Espoir industry.

RECOMMENDATIONS

1. Use of nets of significantly >10 m for over-wintering of salmonids at sites in Bay d'Espoir that have significant current is not economically justifiable for 1st-year salmonids. Net depths of up to 15 m may be useful for improving the economic performance of 2nd-year salmonids but only if maturation is controlled and sources of physiological stress can be identified and remedied.
2. The Bay d'Espoir salmonid aquaculture industry should investigate water-column use by salmonids during the summer months to determine if nets of 15 m depth have any advantage over 10 m nets.
3. The Bay d'Espoir industry should implement a monitoring program for quantification of indicators of early maturation in its aquaculture salmonids. Hormone analyses of the present study suggest frequent blood sampling at times of low environmental stressors may be prove strategic to this goal. Investigation of portable ultrasound equipment for *in situ* measurement of gonad size also should be undertaken.
4. With continuing development of the finfish aquaculture industry in Newfoundland, it becomes increasingly important to assess the level of risk posed by harmful phytoplankton species and determine how improvements to site selection and management might be effected.
5. Use of an underwater video camera and video recorder would be an asset to any future endeavours to understand feeding dynamics and the relation between fluctuating environmental variables and depth distribution of fish in aquaculture nets.

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(NAIA) was instrumental in this regard in co-ordinating views within the industry to identify research priorities that were most significant to most of the industry. We wish to express our appreciation to Bob O'Neill (NAIA) for his many efforts in this regard.

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Table 1: Evaluation of normality and homogeneity of variance between initial and final substrate weights

Species	Year (Years)	Net Depth (m)	Sample Normality (p-values)		Homogeneity of Variance (Levene) p-v
			Shap	Kol	
Sabinea	1999	10	0.659	0.697	0.553
		20	0.515	0.955	0.978
	2000	10	0.659	0.663	0.776
		20	0.659	0.307	0.776
Trem	1999	10	0.148	0.364	0.333
		20	0.193	0.193	0.386
	2000	10	0.033	0.986	0.989
		20	0.233	0.911	0.271

Table 2: Biomass changes during the Matthews river swimming experiment

Species	Year (Years)	Net Depth (m)	Biomass (kg)			Significance
			Shap	Kol	Change (k	p (1) wil (2)
Sabinea	1999	10	2688 (a	2151 (a	709 (a	0.030
		20	6860 (a	5690 (a	2767 (a	0.000
	2000	10	4688 (a	4033 (a	278 (a	0.000
		20	8833 (a	7209 (a	1696 (a	0.000
Trem	1999	10	3870 (a	3686 (a	1233 (a	0.071
		20	10384 (a	8506 (a	3038 (a	0.355
	2000	10	5776 (a	5906 (a	1239 (a	0.000
		20	7333 (a	6548 (a	980 (a	0.000

* Double(d) cells represent significant differences between non-wil and weights

Table 5. Mantel – Haenszel odds ratio tests for differences in survival potential for Atlantic salmon and steelhead trout in 10 m. vs. 25 m. deep aquaculture nets

Species	Year Class	Net Depth (m)	End Salmonid Inventory Data			Odds Ratio Statistics		
			Dead	Alive	Survival (%)	MH ratio	Chi-square	$p <$
Salmon	1999	10	339	665	66			
		25	696	2995	76	1.65	2.174	0.00001
	2000	10	23	1777	99			
		25	28	6472	99			
Trout	1999	10	906	1094	55			
		25	1869	3531	71	1.51	98.85	0.00001
	2000	10	609	1591	80			
		25	999	8001	90			

Table 6. Thermograph records of Bay d'Espoir area water column aquachill conditions during the winters of 2001 and 2002

Location Name	Coordinates		Distance from the Marichorne (km)[1]	Date		Depth (m)	n Records	Temperature < -0.5°	
	Latitude	Longitude		From	To			Low	High
Stucky Cove	47.7075	-56.1633	30	22-Jan-02	9-Feb-02	3.0	9	-0.97	-0.64
Mussels Arm	47.6730	-56.1797	29	20-Jan-02	10-Mar-02	3.0	32	-0.77	-0.61
Gobbin Bay	47.7176	-56.0829	31	15-Feb-01	23-Feb-01	7.6	68	-0.81	-0.64
Crow Island Bay	47.6233	-55.8531		27-Feb-01	6-Mar-01	6.6	54	-0.79	-0.62
Great Collier Bay	47.7263	-56.1790	31	15-Feb-01	24-Feb-01	3.0	60	-1.27	-0.62
Hatcher Arm	47.5905	-56.6105	63	27-Feb-01	5-Mar-01	3.7	197	-1	-0.51
Second Brook Cove	47.8070	-56.1296	39	18-Feb-01	1-Mar-01	3.0	23	-1.19	-0.52
Wild Cove	47.6519	-56.2064	31	24-Feb-01	3-Mar-01	7.6	28	-0.78	-0.62

[1] Water distances from Gobbin Bay, Great Collier Bay and Second Brook Cove are via Little Passage

Table 5. Feeding intensity[1]

Species	Year Class	Net Depth 10 m	Net Depth 25 m
Salmon	1999	1.04	0.38
	2000	0.97	0.41
Trout	1999	0.81	0.48
	2000	0.98	0.60

[1] total food consumed/mean number of fish in cage during interval of experiment (kg•fish^{-1})

Table 6. Instantaneous rates for growth (G), mortality (Z) and biomass increment (R) for the Matchams salmonid over-wintering experiment.

Species	Sample	Year Class	Cage Depth (m)	Mean Individual Live Weight (g)	Mean Individual Fork Length (cm)	Number	G[1]	Z[1]	R[1]
Salmon	Start	2000	10	914.0	40.3	1800			
	End			1082.1	42.6	1777	0.0011	0.0001	0.0010
	Start	1999	10	2948.6	63.1	1000			
	End			3254.5	65.9	661	0.0006	0.0027	-0.0021
	Start	2000	25	914.0	40.3	4500			
	End			1115.0	44.4	4472	0.0016	0.0000	0.0015
	Start	1999	25	2926.4	63.1	2891			
	End			2593.3	62.4	2195	-0.0008	0.0018	-0.0026
Steelhead	Start	2000	10	1390.3	42.3	2000			
	End			1826.7	44.7	1591	0.0018	0.0015	0.0003
	Start	1999	10	2020.0	47.5	2000			
	End			2231.5	48.4	1094	0.0011	0.0066	-0.0055
	Start	2000	25	1429.0	42.2	5000			
	End			1636.7	43.9	4001	0.0009	0.0015	-0.0006
	Start	1999	25	2118.7	48.2	5000			
	End			2248.1	48.3	3531	0.0006	0.0038	-0.0031

[1] daily instantaneous rates

Table 7. Percent of DST depth records from 19 tagged specimens by 2.5 m depth categories.

Species	Year Class	Net Depth	Tag #	Percentage of observations at depth (m.)							
				<2.5	2.5 - <5	5 - <7.5	7.5 - <10	10 - <12.5	12.5 - <15	15- <17.5	≥ 17.5
	1999	10		No DSTs applied to 2nd-year salmon (i.e., grilse)							
	1999	25									
			38	58.5	37.1	4.2	0.1				
			62	31.6	65.2	3.0	0.2				
			71	16.0	67.5	16.0	0.6				
Salmon	2000	10	74	74.2	24.8	0.2					
			39	59.0	37.9	2.7	0.3	0.1			
		25	46	36.3	57.9	5.5	0.4				
			61	38.4	54.0	5.0	1.6	0.6	0.8	0.2	0.3
			69	21.3	67.0	6.4	2.3	1.7	0.8	0.2	0.3
		10		No DSTs recovered from 2nd-year steelhead in the 10 m net							
	1999	25	68	58.9	37.7	1.9					
			73	3.5	35.8	55.4	4.9	0.3	0.1		
			36	70.0	28.1	2.0					
		10	51a	82.7	16.5	0.7					
			51b	49.8	45.1	5.1					
Steelhead			52	78.8	15.1	5.8	0.3				
			41	50.4	10.2	11.2	18.1	7.9	1.8	0.3	0.3
	2000		42	92.3	7.2	0.1					
			43	14.7	5.1	5.8	14.1	23.7	21.3	9.9	5.5
		25	45a	85.5	14.3	0.2					
			45b	24.9	66.3	2.8	2.7	2.6	0.6	0.1	

Table 8. Magnitude of discrepancies noted among DSTs from the same depth at the same time.

Variable Category	Number of Records	% of Observations
Temperature		
<0.5 C	1110	92.7
>= 0.5 and <1.0 C	66	5.5
>= 1.0 and < 1.5 C	17	1.4
>= 1.5 and <= 2.0 C	5	0.4
Salinity		
<2.0 ppt	979	96.6
>=2.0 and < 4.0 ppt	27	2.7
>= 4.0 and < 6.0 ppt	5	0.5
>= 6.0 ppt	2	0.2

Table 9. Percentage breakdown of frequency of DST salinity and temperature data for the Matchums salmonids.

	Salinity (‰)				Temperature (°C)		
Species	< 10	≥ 10.0 and < 20.0	≥ 20.0 and < 30.0	≥ 30.0	< -0.5	≥ -0.5 and < 0.0	≥ 0.0
Atlantic salmon	0.02	0.08	70.3	29.6	0.05	0.75	99.2
Steelhead	0.07	0.08	53.6	46.3	0.009	0.64	99.4

Table 10. Blood parameters for cage depth study.

Specimen	Osmolality (mmol/kg)			Cortisol (µg/dL)			Glucose (mg/dL)			17β-Estradiol (pg/ml)		
	Mean ± SE	Min	Max	Mean ± SE	Min	Max	Mean ± SE	Min	Max	Mean ± SE	Min	Max
SH2-10*	474.0 ± 32.9	314.0	593.0	18.5 ± 5.3	0.2	37.8	117.8 ± 19.0	68.8	248.8	19.11 ± 4.87	0.73	49.29
SH2-25	505.7 ± 15.4	432.0	569.0	16.9 ± 4.0	1.3	37.7	154.6 ± 35.5	62.4	378.0	38.21 ± 7.90	5.57	79.09
SH1-10	346.9 ± 3.3	336.0	362.0	2.7 ± 0.8	0.0	7.2	79.5 ± 8.1	62.0	131.0	87.05 ± 16.98	34.82	173.02
SH1-25	350.1 ± 4.8	335.0	377.0	5.8 ± 1.2	1.1	10.5	70.1 ± 3.2	57.0	82.0	102.73 ± 20.94	50.69	199.33
AS1-10**	356.2 ± 3.2	346.0	376.0	9.2 ± 2.2	0.0	19.4	81.5 ± 3.3	69.0	97.0	NA	NA	NA
AS1-25	357.0 ± 2.8	347.0	375.0	9.4 ± 1.5	3.0	16.8	74.8 ± 2.5	60.0	91.0	NA	NA	NA
AS2-10	361.0 ± 2.8	348.0	377.0	12.6 ± 2.6	5.3	27.9	81.1 ± 2.7	72.0	95.0	NA	NA	NA
AS2-25	350.3 ± 2.9	338.0	362.0	2.9 ± 1.2	0.0	8.0	69.3 ± 5.9	55.0	109.0	NA	NA	NA

* SH2-10: SH = Steelhead, (2-10) first number indicates marine year and second number is cage depth in metres.
** AS = Atlantic salmon.

Figure 1. Bay d'Espoir study area and site location.

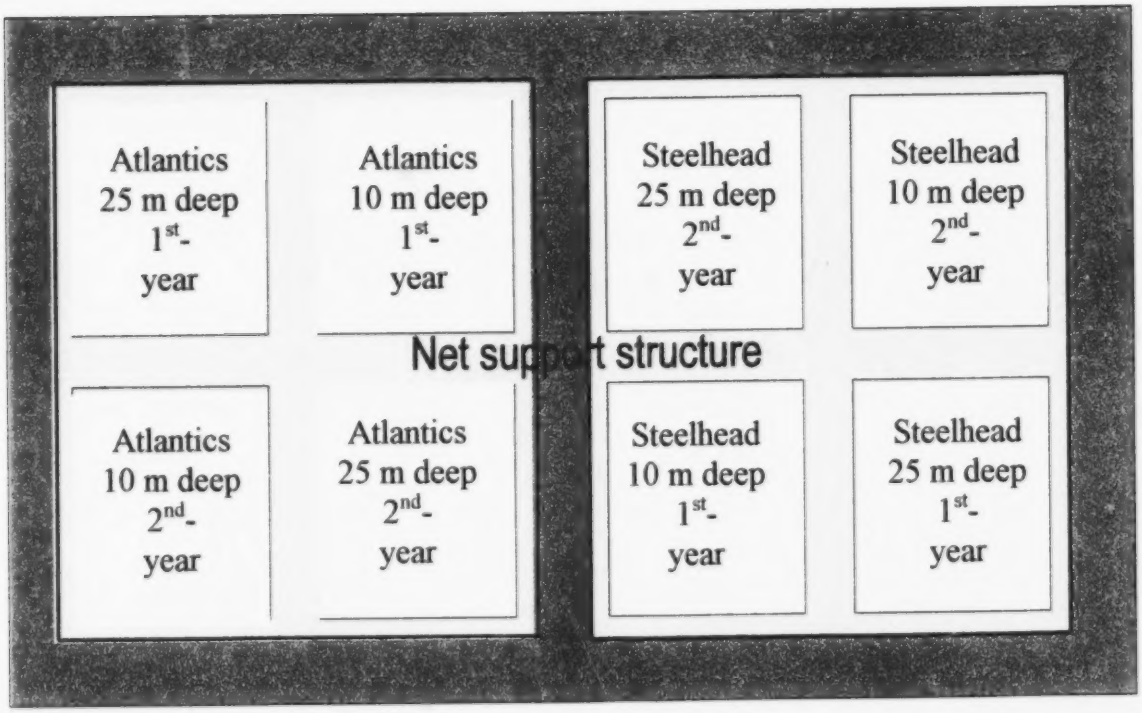

Atlantics 25 m deep 1st-year
Atlantics 10 m deep 1st-year
Steelhead 25 m deep 2nd-year
Steelhead 10 m deep 2nd-year
Net support structure
Atlantics 10 m deep 2nd-year
Atlantics 25 m deep 2nd-year
Steelhead 10 m deep 1st-year
Steelhead 25 m deep 1st-year

Figure 2. Experimental design to evaluate optimal net depth for over-wintering of salmonids in Bay d'Espoir.

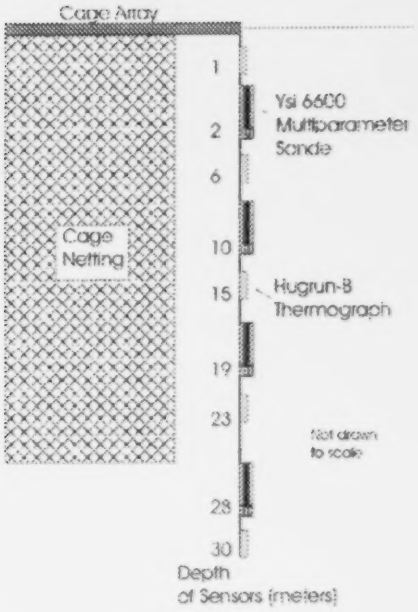


Figure 3. Water-column monitoring sonde and recording-thermograph array at the Matchums over-wintering site.

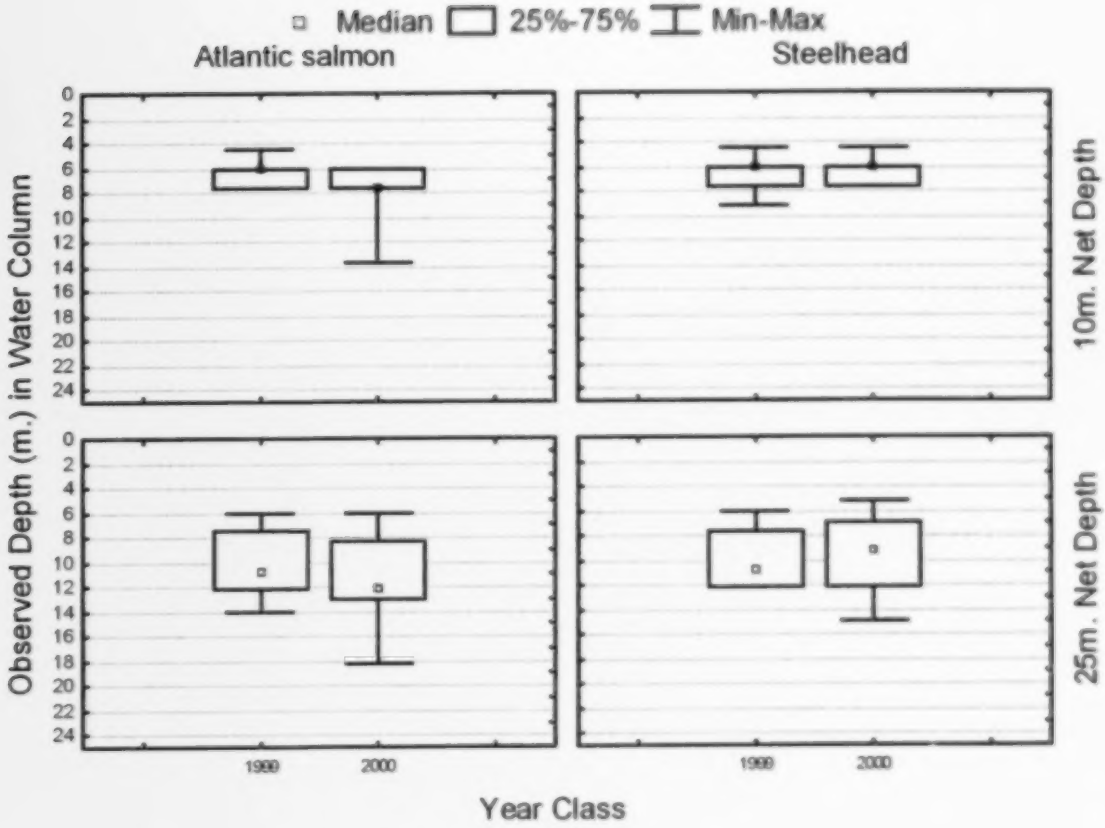


Figure 4. Diver observations of vertical distribution of most of the fish in the water column.

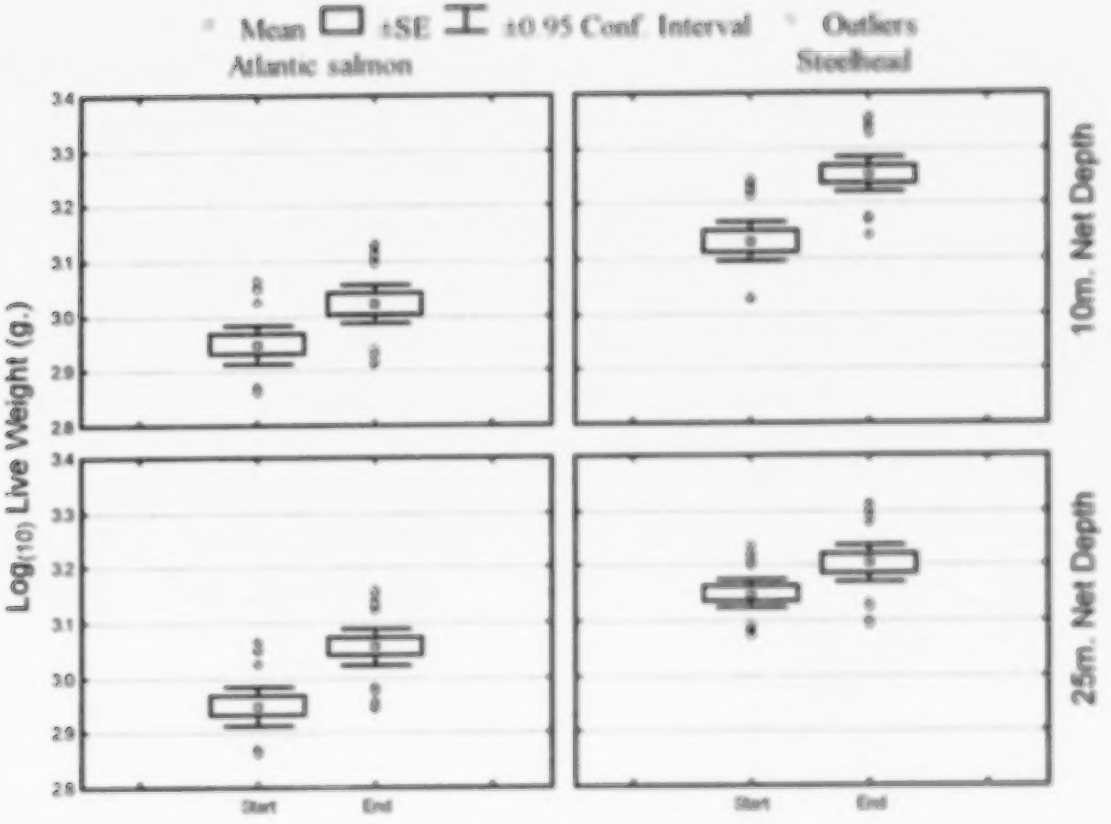


Figure 5. 1st-year, Atlantic salmon and steelhead sampling statistics (n = 35 for each sample).

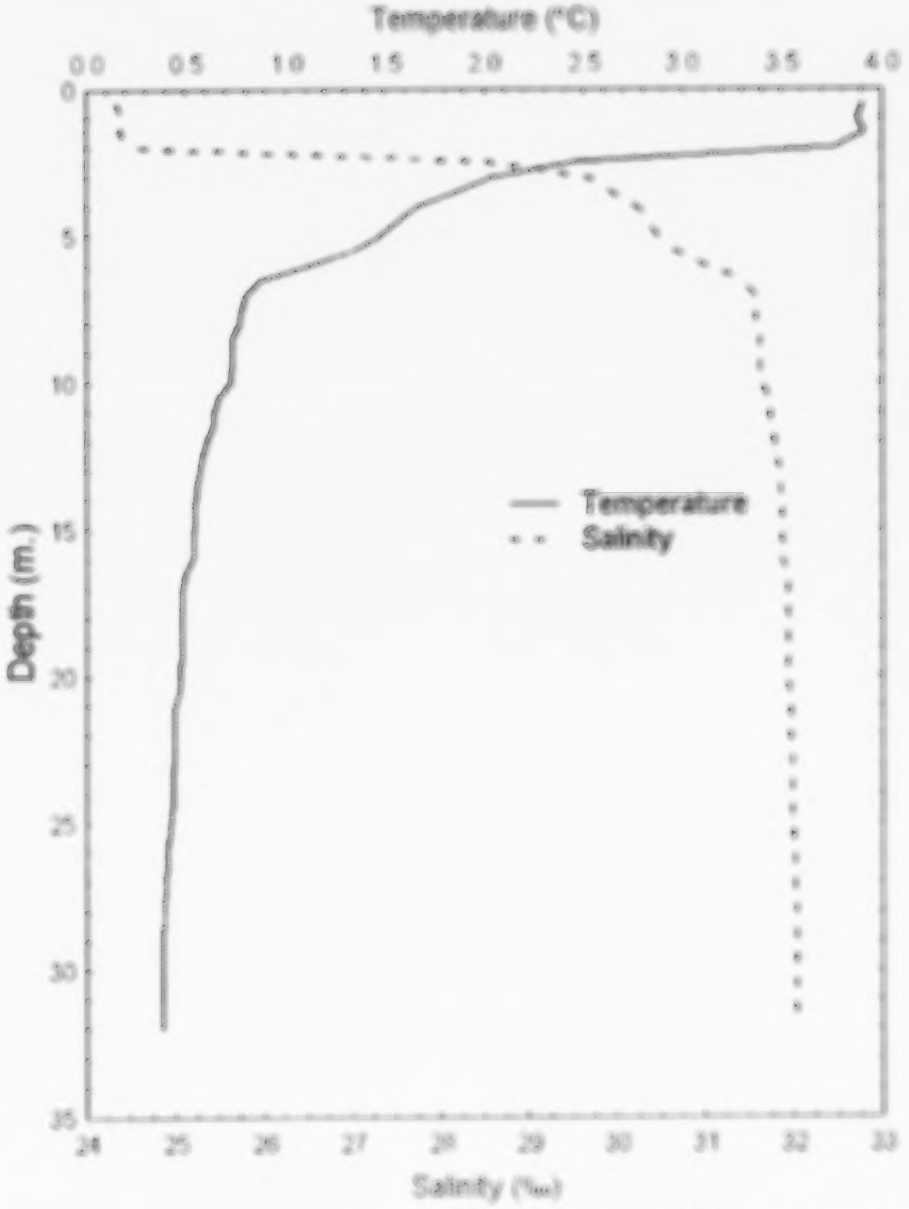


Figure 6. CTD profile of water column temperature and salinity characteristics for the Matchums (April 16/02)

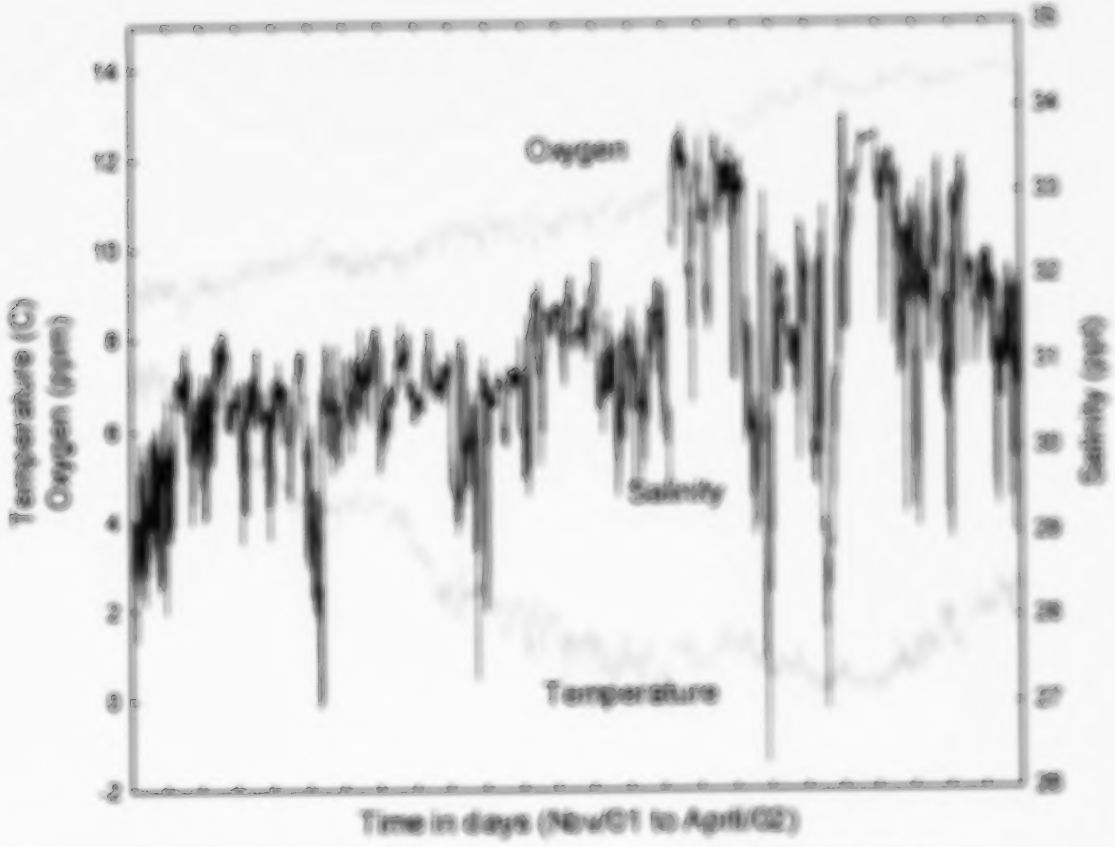


Figure 7. The Matchums, temperature, oxygen and salinity variability at 3 m.

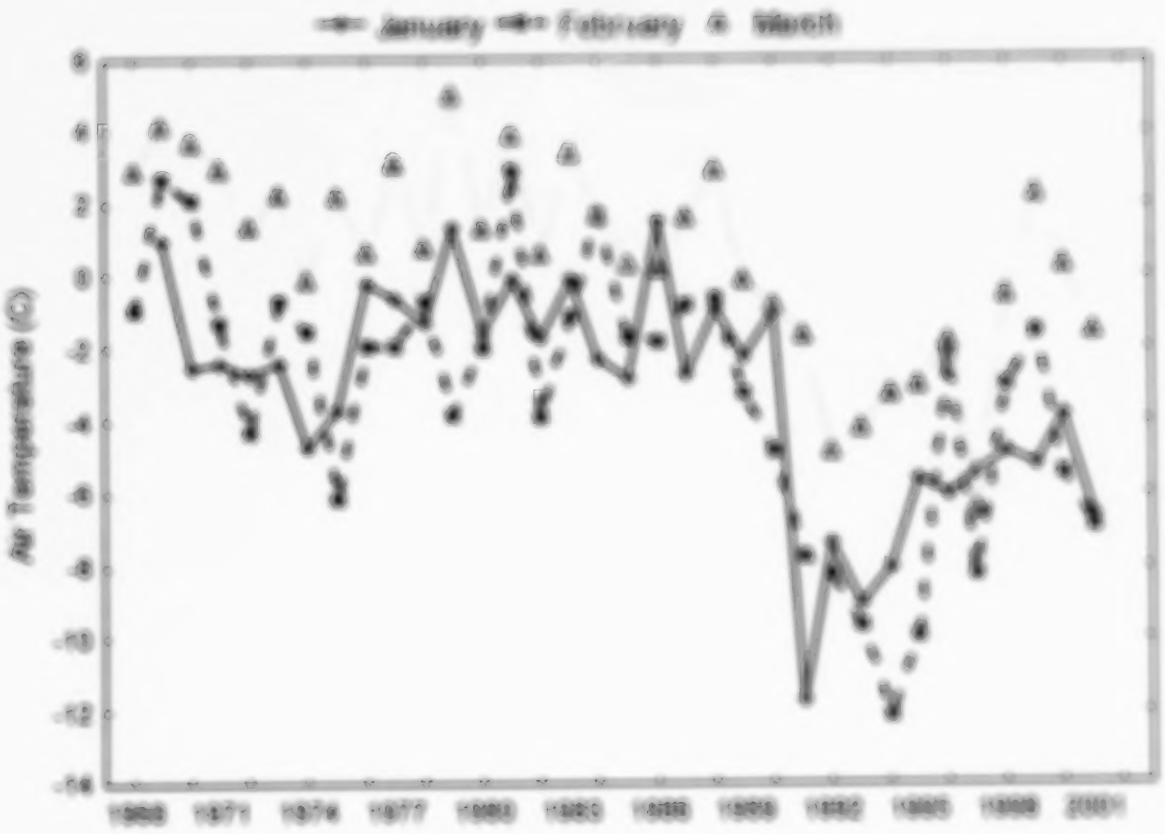


Figure 6. Historical winter air temperatures for Bay d'Espoir Hydroelectric Generating Station (40 km from the Matchbums aquaculture site).

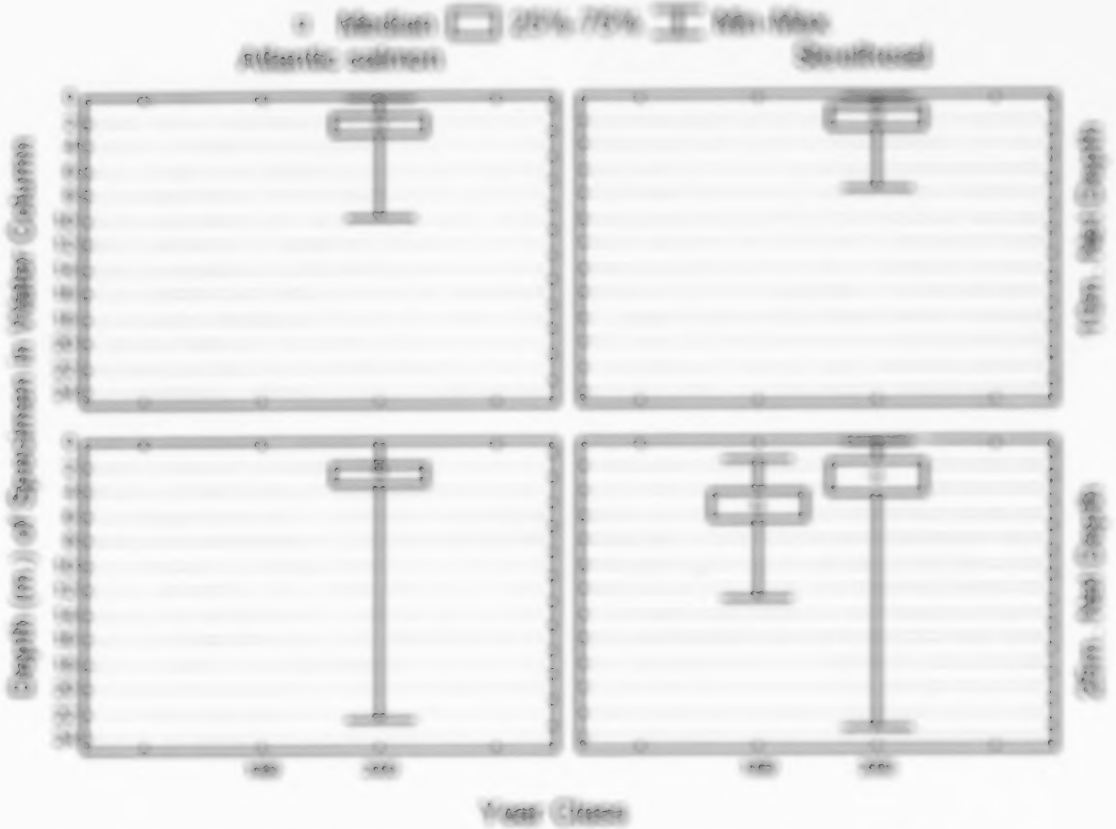


Figure 9 DSD records of winter water column run by approximate schematic

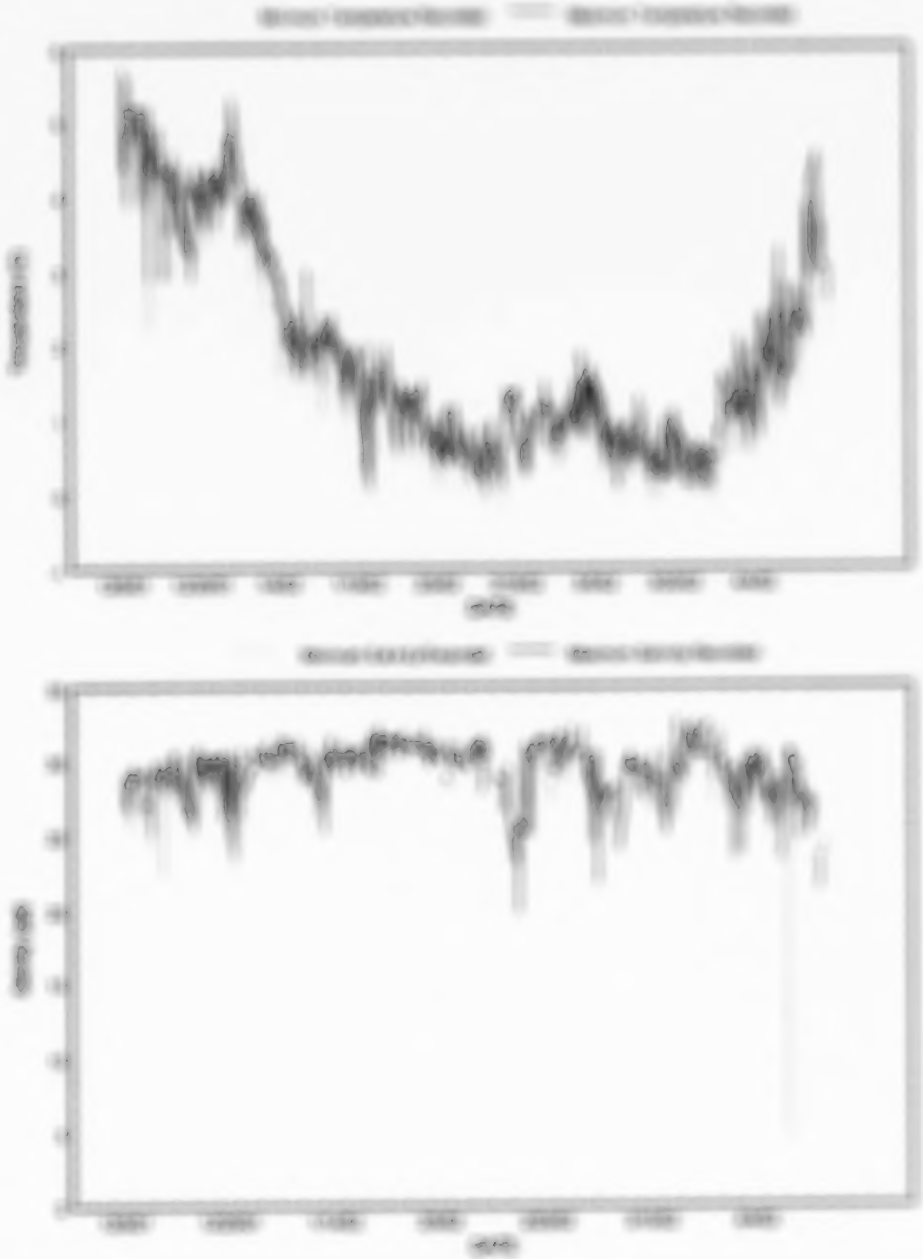


Figure (1). Difference among TET measurements recorded at a common depth and a common time.

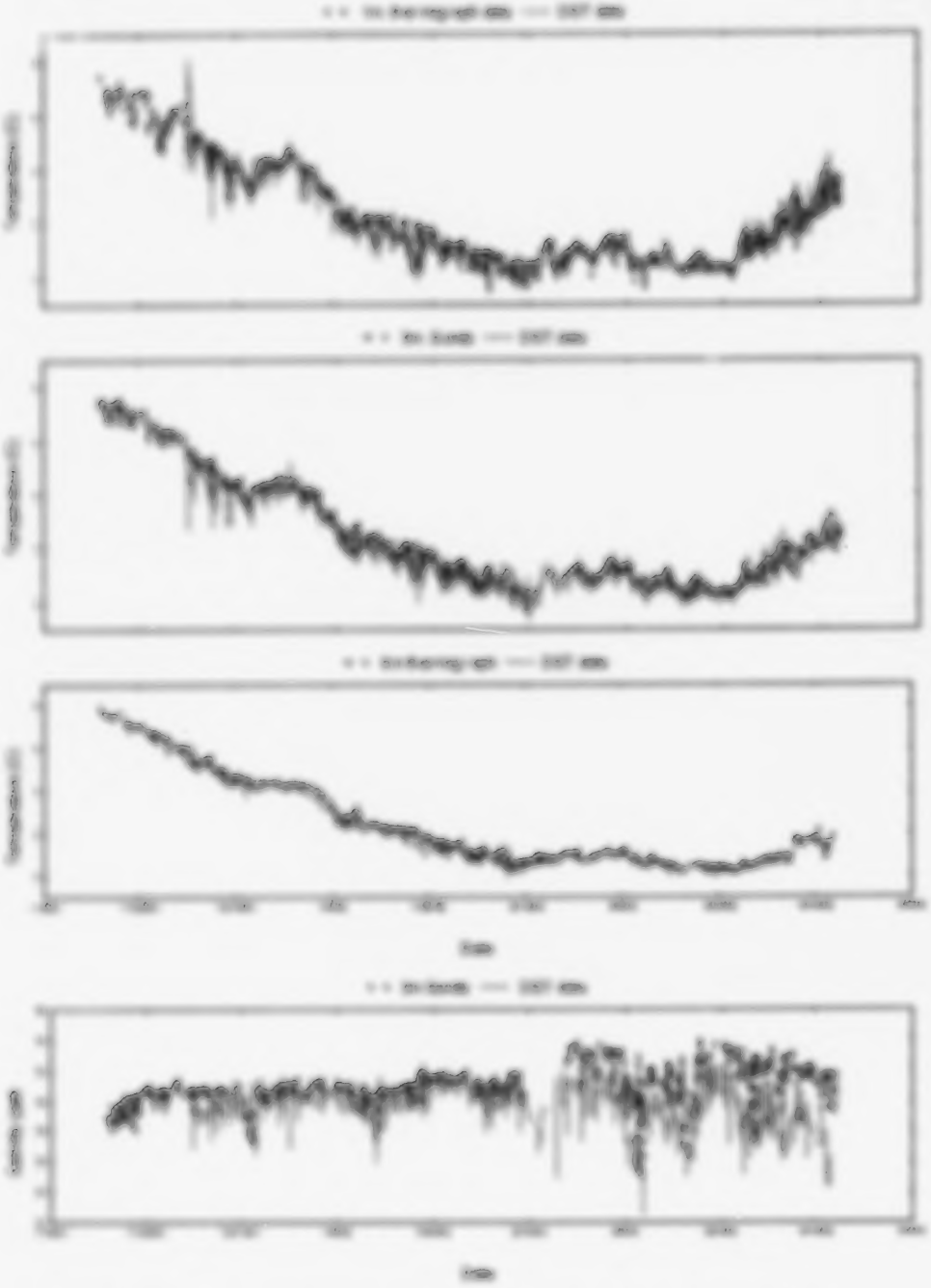
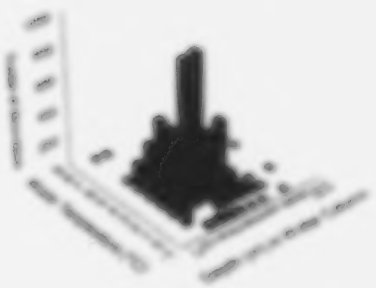
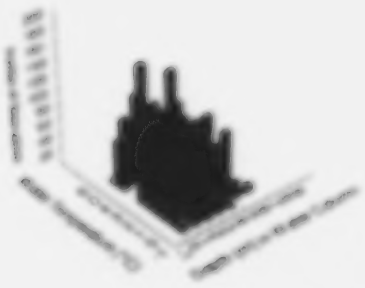


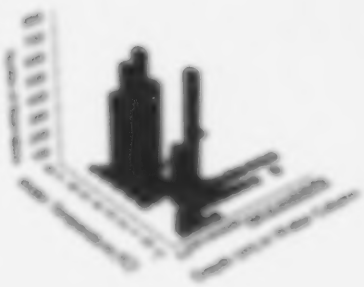
Figure 11. Comparison of water temperature and salinity records for CTDs with data of fixed-monitoring gear.



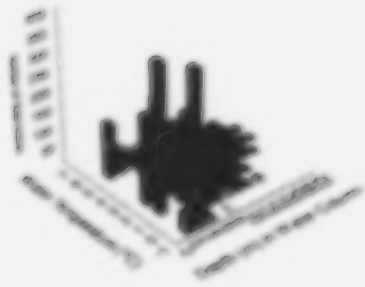
1st-year Atlantic Salmon, 10m Net



1st-year Steelhead, 10m Net



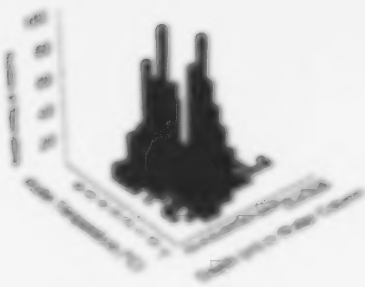
1st-year Atlantic Salmon, 25m Net



1st-year Steelhead, 25m Net

Observed frequency (%) of depth distribution for the Matchums salmonids.

Species	Depth	
	<10m	>10m
Atlantic salmon	99.2	0.8
Steelhead	87.6	12.4



2nd-year Steelhead

Figure 12. DST records of water depths and temperatures experienced by the Matchums salmonids during January through March, 2002.

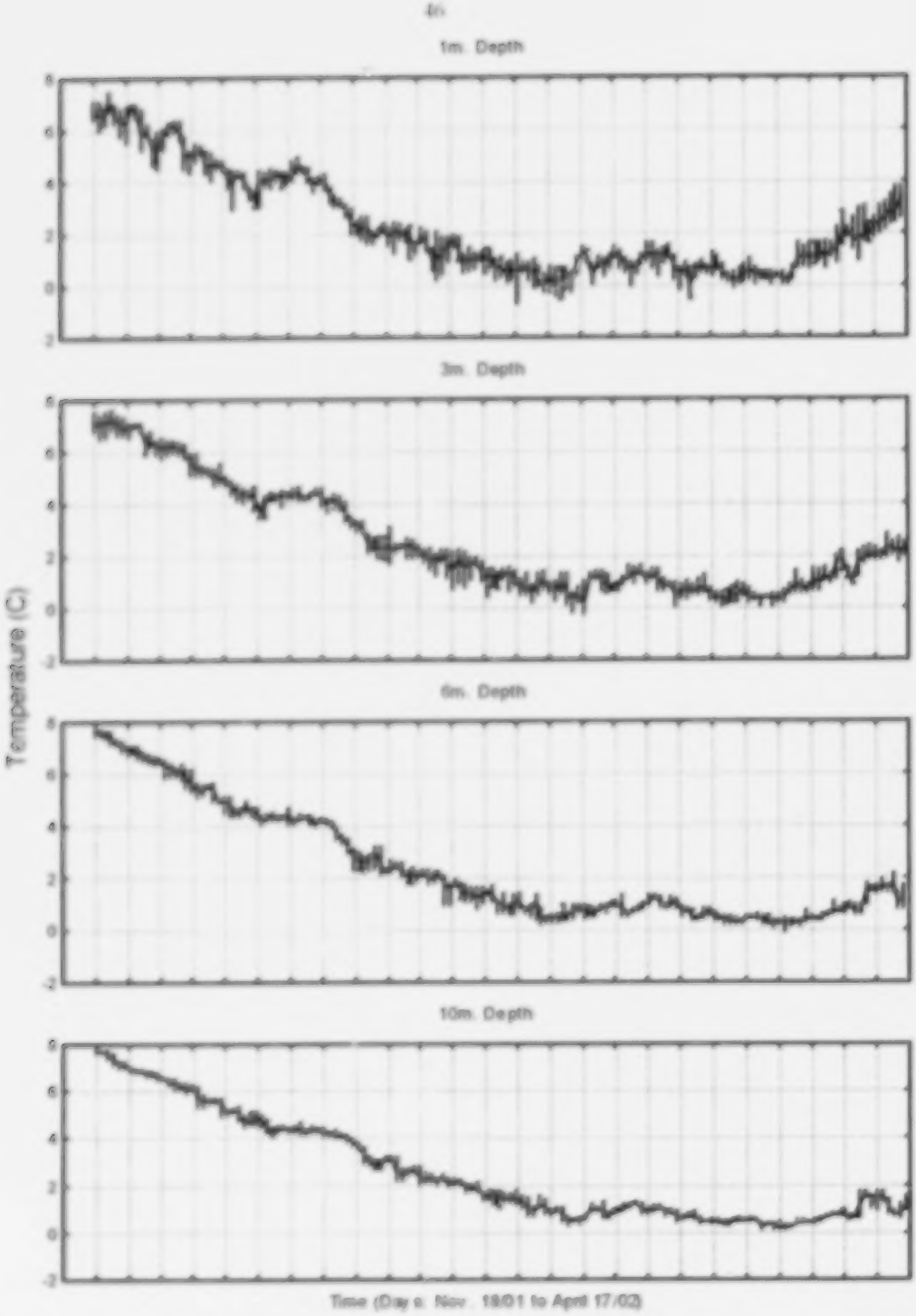


Figure 13. Water-column temperature records for fixed monitoring apparatus. Vertical bars represent daily temperature range.

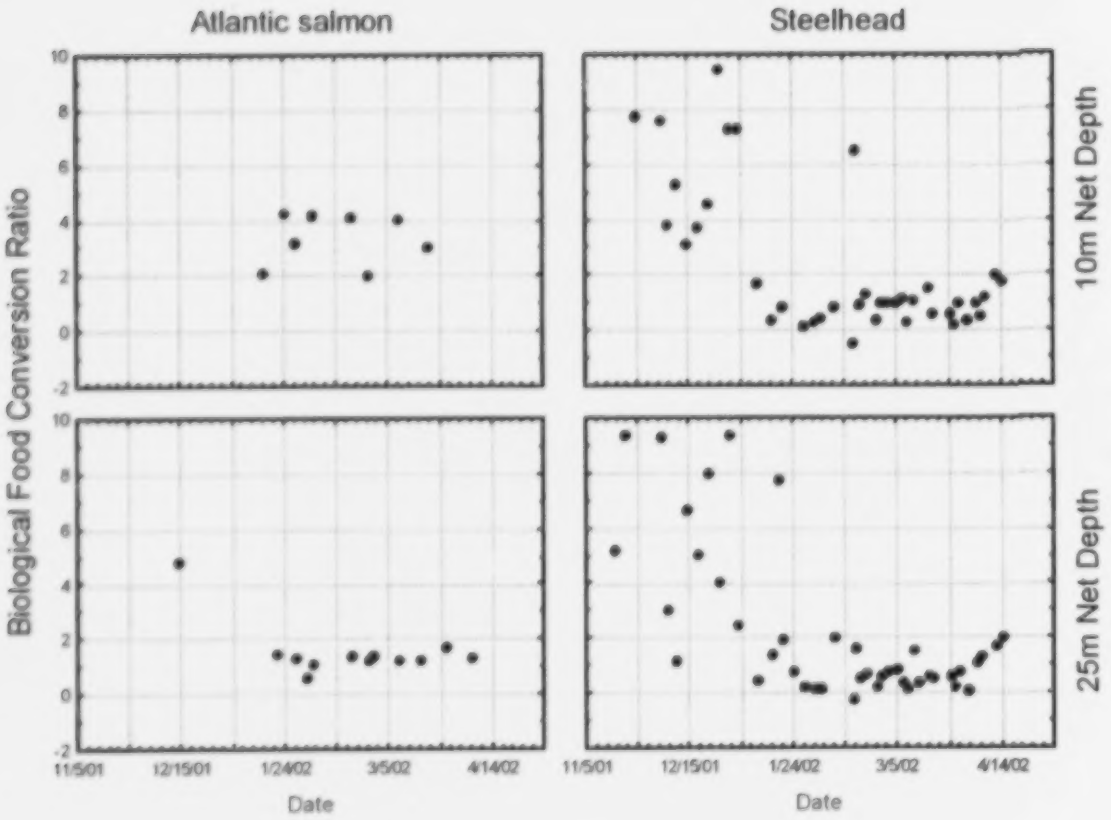


Figure 14. Biological food-conversion ratios over time.

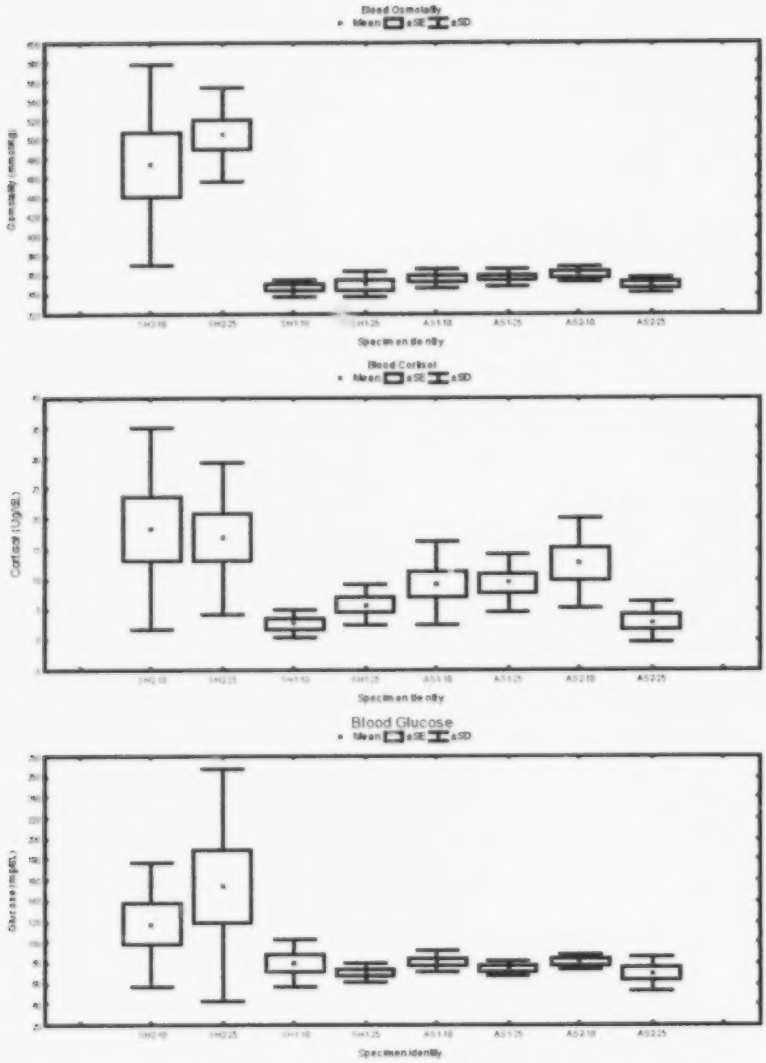


Figure 15. Blood chemistry analyses of steelhead (SH) and Atlantic salmon (AS).

Appendix 1. Thermograph summary, the Matchums: winter 2001/02.

Thermograph Unit	Month	Thermal Units[1]	Water Temperature (°C at depth)		
			Mean	Min	Max
Hugrun,	November	84.7	6.35	4.6	7.5
MS110	December	141.3	4.56	3.0	6.4
	January	57.0	1.84	0.3	3.8
1 m depth	February	17.9	0.64	-0.7	1.5
	March	19.8	0.64	-0.6	1.7
	April	36.7	2.05	0.7	3.8
	Σ Thermal Units	357.4			
YSI, 6600	November	96.1	6.86	5.8	7.6
	December	146.7	4.73	3.5	6.4
3 m depth	January	63.4	2.05	0.5	4.1
	February	23.0	0.82	-0.2	1.4
	March	20.5	0.68	0.1	1.6
	April	30.0	1.67	0.6	3.0
	Σ Thermal Units	379.7			
Hugrun,	November	94.1	7.06	6.4	8.0
MS110	December	151.0	4.87	4.0	6.6
	January	69.5	2.24	0.8	4.2
6 m depth	February	21.9	0.78	0.3	1.4
	March	17.2	0.55	-0.1	1.3
	April	19.7	1.11	0.4	2.2
	Σ Thermal Units	373.4			
YSI, 6600	November	99.4	7.10	6.5	7.9
	December	154.4	4.98	4.0	6.6
10 m depth	January	74.2	2.39	1.1	4.1
	February	24.9	0.89	0.3	1.6
	March	18.0	0.60	0.1	1.3
	April	17.2	0.96	0.4	1.9
	Σ Thermal Units	388.1			
Hugrun,	November	95.0	7.12	6.2	8.1
MS110	December	159.9	5.16	4.2	6.7
	January	80.9	2.61	1.4	4.2
15 m depth	February	27.3	0.98	0.5	1.7
	March	21.7	0.70	0.3	1.4
	April	15.8	0.89	0.5	1.7
	Σ Thermal Units	400.6			

Appendix 1 (Cont'd.)

Thermograph Unit	Month	Thermal Units[1]	Water Temperature (°C at depth)		
			Mean	Min	Max
YSI, 6600	November	89.7	6.41	5.9	7.4
	December	142.1	4.58	3.5	6.0
19 m depth	January	61.9	2.00	0.8	3.6
	February	12.1	0.43	0.0	1.0
	March	Sonde function erratic following redeployment in February			
	April				
Hugrun,	November	90.2	6.76	6.1	7.9
MS110	December	156.2	5.04	4.0	6.4
	January	77.0	2.48	1.2	4.1
23 m depth	February	10.8	0.91	0.5	1.4
	March	Thermograph unit ceased functioning on February 12.			
	April				
YSI, 6600	November	93.0	6.64	5.7	7.9
	December	156.6	5.05	3.7	6.4
28 m depth	January	78.9	2.55	1.3	4.1
	February	27.3	0.97	0.6	1.5
	March	15.4	0.51	0.1	1.2
	April	9.5	0.53	0.4	1.1
	Σ Thermal Units	380.7			
Hugrun,	November	89.5	6.71	5.5	8.0
MS110	December	158.0	5.10	3.6	6.5
	January	79.4	2.56	1.3	4.2
30 m depth	February	23.3	0.83	0.3	1.5
	March	14.8	0.48	0.1	1.2
	April	2.3	0.49	0.4	0.7
	Σ Thermal Units	367.3			

[1] mean daily temperature x number of days. Thermal units accumulated over only 13d in November and 18d in April.

Appendix 2. Other salmonid performance measures for the Matchums, November 2001 through April, 2002.

Species	Net Depth	Month	Year	Mean Weight		Total Feed	Biomass			Econ. FCR[1]	Mortality Biomass	Bio. FCR	Therm. Units	GF3
				Start	End		Start	End	Added					
		Nov.	2001	914.0	929.3	175.0	1644.3	1670.1	25.8	6.78	0.9	6.55		
		Dec.	2001	930.3	961.6	414.0	1672.7	1724.2	51.5	8.04			113.9	0.95
		Jan.	2002	962.7	995.1	367.0	1726.1	1780.2	54.1	6.78	3.9	6.32	56.7	1.93
	10	Feb.	2002	996.2	1026.3	251.3	1782.2	1826.8	44.6	5.63	9.1	4.68	20.7	4.81
		Mar.	2002	1027.4	1062.0	344.0	1828.8	1887.2	58.4	5.89	3.1	5.59	23.3	4.80
		Apr.	2002	1063.2	1082.1	175.0	1889.3	1923.0	33.7	5.19			38.4	1.57
		Nov.	2001	914.0	936.0	200.0	4113.0	4210.1	97.1	2.06	1.9	2.02		
Salmon		Dec.	2001	937.5	983.2	494.0	4216.8	4426.2	209.4	2.36	1.0	2.35	110.9	1.41
		Jan.	2002	984.7	1032.7	360.0	4433.3	4646.2	212.9	1.69	3.1	1.67	53.9	2.95
	25	Feb.	2002	1034.4	1079.6	212.8	4653.6	4838.8	185.2	1.15	18.0	1.05	19.0	7.66
		Mar.	2002	1081.3	1134.0	361.5	4846.5	5073.6	227.1	1.59	8.9	1.53	19.6	8.36
		Apr.	2002	1135.8	1165.0	212.5	5081.6	5209.9	128.3	1.66	2.3	1.63	35.1	2.53
		Nov.	2001	1390.3	1428.0	282.0	2776.4	2850.3	73.9	3.82	1.4	3.74		
		Dec.	2001	1430.6	1509.2	637.5	2855.4	2995.8	140.4	4.54	16.1	4.07	114.9	1.76
		Jan.	2002	1511.9	1595.0	360.0	3001.1	3032.2	31.1	11.58	131.8	2.21	49.5	4.18
	10	Feb.	2002	1597.9	1676.7	175.0	3037.6	2907.5	-130.1	na	271.7	1.24	6.9	27.42
		Mar.	2002	1679.7	1772.1	212.5	2912.7	2909.8	-2.9	na	159.2	1.36	18.2	11.73
Trout		Apr.	2002	1775.3	1826.7	150.0	2915.0	2906.2	-8.8	na	91.2	1.82	32.3	3.58
		Nov.	2001	1429.0	1448.1	556.0	7133.6	7216.1	82.5	6.74	12.9	5.83	13.5	3.69
		Dec.	2001	1449.4	1488.5	1012.0	7222.5	7307.1	84.6	11.96	108.3	5.25	118.7	0.85
		Jan.	2002	1489.8	1530.0	521.3	7315.5	7273.6	-41.9	na	234.8	2.70	63.1	1.61
	25	Feb.	2002	1531.4	1568.5	237.5	7280.0	6918.5	-361.5	na	529.8	1.41	18.7	4.93
		Mar.	2002	1569.9	1612.2	237.5	6924.6	6698.6	-226.0	na	408.0	1.31	17.7	5.86
		Apr.	2002	1613.6	1636.7	212.5	6548.3	6548.3	-585.3	na	249.4	-0.63	31.1	1.79

[1] Negative values of the Economic FCR are eliminated as not applicable.

Appendix 3: Salmonid specimen measurements and blood analyses.

February 2002 Samples:

Fish ID	Tag	Length (cm)	Weight (g)	Glucose (mg/dL)	Osmolality (mmol/Kg)	Total Protein (g/dL)	Cortisol (Ug/dL)	Estradiol (pg/ml)	Notes
SH2-10-1	N	47.50	1888.00	76.80	402.00	4.04	1.60	12.84	
SH2-10-2	N	50.50	2677.00	248.80	568.00	6.82	35.09	19.31	
SH2-10-3	N	44.00	1973.00	82.40	363.00	3.28	2.56	36.73	
SH2-10-4	N	46.00	2050.00	180.00	586.00	6.78	37.61	30.75	
SH2-10-5	N	52.50	2705.00	159.60	554.00	6.21	35.42	5.78	
SH2-10-6	N	45.00	1629.00	87.60	535.00	5.45	22.26	11.00	
SH2-10-7	N	49.50	2178.00	121.20	593.00	6.97	37.80	49.29	
SH2-10-8	N	43.00	1754.00	72.80	437.00	10.59	6.69	18.06	
SH2-10-9	N	44.00	1423.00	68.80	314.00	0.20	0.19	6.61	
SH2-10-10	N	41.00	1453.00	80.40	388.00	1.94	5.48	0.73	
SH2-25-1	N	47.00	1602.00	103.60	455.00	3.73	15.65	57.48	
SH2-25-2	N	37.50	1026.00	62.80	477.00	1.78	4.66	15.86	
SH2-25-3	N	44.50	1692.00	146.80	517.00	6.10	19.39	79.09	
SH2-25-4	N	44.00	1782.00	87.60	554.00	3.81	25.95	15.70	
SH2-25-5	N	47.50	2476.00	118.00	541.00	4.51	20.94	49.69	
SH2-25-6	N	43.50	2172.00	378.00	529.00	7.70	32.20	37.20	
SH2-25-7	N	39.00	1109.00	62.40	569.00	3.44	37.71	31.11	
SH2-25-8	N	41.50	1414.00	85.60	447.00	3.56	1.34	52.18	
SH2-25-9	N	55.00	1992.00	337.20	536.00	2.81	7.98	5.57	
SH2-25-10	N	45.50	1379.00	163.60	432.00	0.88	3.26	Out	

April 2002 Samples:

Fish ID	Tag	Length (cm)	Weight (g)	Liver weight (g)	Hepato-Somatic Index	Osmolality (mmol/Kg)	Total Protein (g/dL)	Cortisol (Ug/dL)	Estradiol (pg/ml)	Notes
SH1-10-1	Y	46.50	1880.00	50.00	2.13	339.00	1.65	0.03	34.82	
SH1-10-2	Y	46.50	2265.00	40.00	1.77	341.00	2.15	2.07	96.56	
SH1-10-3	Y	44.50	1670.00	35.00	2.10	352.00	1.81	2.18	57.71	
SH1-10-4	Y	45.50	1855.00	35.00	1.89	342.00	3.20	3.09	82.44	
SH1-10-5	N	41.60	1620.00	70.00	4.32	336.00	8.49	0.80	8874.92	Gravid female
SH1-10-6	N	47.50	2030.00	40.00	1.97	346.00	4.86	1.24	173.02	
SH1-10-7	N	46.60	2040.00	35.00	1.72	357.00	4.88	7.18	104.58	
SH1-10-8	N	46.60	1670.00	25.00	1.50	362.00	4.66	4.99	60.24	
SH1-25-1	Y	46.50	1965.00	60.00	3.05	344.00	4.44	2.82	6396.28	Gravid female
Sh1-25-2	Y	44.70	1605.00	40.00	2.49	362.00	2.74	4.58	Out	
SH1-25-3	N	42.80	1235.00	20.00	1.62	335.00	5.05	1.12	50.69	
SH1-25-4	N	47.30	1955.00	30.00	1.53	340.00	5.29	10.24	110.47	
SH1-25-5	N	42.30	1415.00	25.00	1.77	351.00	5.09	4.96	79.82	
SH1-25-6	N	42.30	1585.00	20.00	1.26	377.00	4.94	5.34	80.82	
SH1-25-7	N	42.30	1420.00	20.00	1.41	349.00	4.14	10.52	199.33	
SH1-25-8	N	45.20	1715.00	30.00	1.75	343.00	4.27	6.85	95.25	

Appendix 3a. (Cont'd.)

Fish ID	Tag	Length (cm)	Weight (g)	Liver weight (g)	Hepato-Somatic Index	Chromidity (mmol/kg)	Total Protein (g/dL)	Cortisol (U/g/dL)	Estradiol (pg/ml)	Notes
AS1-10-1	Y	44.00	1030.00	35.00	3.40	364.00	0.72	3.97	Out	
AS1-10-2	Y	45.60	1090.00	25.00	2.29	340.00	2.57	0.00	Out	
AS1-10-3	Y	41.80	900.00	25.00	2.78	349.00	2.85	0.24	Out	
AS1-10-4	Y	50.90	1700.00	45.00	2.65	376.00	3.37	3.86	Out	
AS1-10-5	Y	41.80	930.00		0.00	360.00	3.79	10.30	Out	Liver
AS1-10-6	N	44.90	1185.00	20.00	1.75	349.00	8.25	19.40	Out	
AS1-10-7	N	47.90	1340.00	20.00	1.49	355.00	9.20	16.89	Out	
AS1-10-8	N	38.50	755.00	15.00	1.99	358.00	5.16	0.99	Out	Liver
AS1-10-9	N	45.60	1105.00	20.00	1.81	349.00	8.90	16.07	Out	
AS1-10-10	N	39.40	855.00	15.00	1.75	346.00	8.79	16.49	Out	
AS1-25-1	Y	46.10	1270.00	30.00	2.36	358.00	3.46	2.99	Out	
As1-25-2	Y	46.80	1275.00	35.00	2.75	347.00	3.40	3.08	Out	
AS1-25-3	Y	47.30	1435.00	35.00	2.44	366.00	6.29	9.29	Out	
AS1-25-4	Y	47.60	1410.00	45.00	3.19	375.00	3.13	0.66	Out	
AS1-25-5	Y	45.30	1010.00	25.00	2.48	360.00	3.29	12.33	Out	Severe cellulitis
AS1-25-6	N	47.60	1350.00	25.00	1.85	348.00	6.16	10.16	Out	
AS1-25-7	N	44.50	1265.00	25.00	1.98	353.00	3.96	6.33	1.86	
AS1-25-8	N	41.70	880.00	20.00	2.27	357.00	6.86	15.89	Out	Skin in bad shape
AS1-25-9	N	45.60	1225.00	20.00	1.63	362.00	3.42	0.41	Out	
AS1-25-10	N	46.50	1235.00	20.00	1.62	358.00	3.31	16.77	7.59	
AS2-10-1	N	74.60	4755.00	65.00	1.37	362.00	3.01	3.32	Out	
AS2-10-2	N	65.80	3310.00	75.00	2.27	348.00	6.97	6.11	Out	
AS2-10-3	N	62.50	2980.00	35.00	1.86	360.00	6.86	6.91	Out	
AS2-10-4	N	68.00	3130.00	65.00	2.08	359.00	3.35	9.52	Out	
AS2-10-5	N	65.50	2810.00	85.00	3.03	358.00	3.36	16.26	Out	
AS2-10-6	N	66.00	3010.00	60.00	1.99	377.00	6.86	27.88	Out	
AS2-10-7	N	75.90	4880.00	65.00	1.33	363.00	3.05	16.63	Out	
AS2-10-8	N	66.00	2960.00	50.00	1.68	361.00	3.46	15.89	Out	
AS2-25-1	N	59.80	2165.00	85.00	2.08	358.00	3.86	0.09	Out	
AS2-25-2	N	65.80	3175.00	65.00	2.09	362.00	6.63	0.00	Out	
AS2-25-3	N	66.00	2650.00	60.00	2.28	345.00	3.40	0.00	Out	
AS2-25-4	N	65.90	2710.00	60.00	2.21	352.00	3.09	3.28	Out	
AS2-25-5	N	62.00	2495.00	85.00	3.53	355.00	6.96	3.38	Out	
AS2-25-6	N	63.80	2280.00	50.00	2.23	350.00	3.70	6.52	Out	
AS2-25-7	N	57.60	1670.00	30.00	1.80	366.00	3.33	0.06	Out	
AS2-25-8	N	60.00	1975.00	85.00	2.28	360.00	3.33	7.99	Out	

Appendix 18. Salmonid blood chemistry

Database (Erlang ETAB= Enterologen, April 2002).

Fish ID	Glucose (mg/dL)	Sodium (mmol/L)	Potassium (mmol/L)	TCO_2 (mmol/L)	Ionized Calcium (mmol/L)	Hematocrit (% PVC)	Hb (g/dL)	pH	PCO_2 (mmHg)	PO_2 (mmHg)	HCO_3 (mmol/L)	BEecf (mmol/L)	sO_2 (%)
1001-10-1	83.00	168.00	<2	8.00	1.60	26.00	8.80	7.02	55.90	18.00	8.00	-25.00	60.00
1001-10-2	67.00	175.00	3.20	8.00	1.59	29.00	9.90	7.03	56.60	21.00	8.00	-22.00	85.00
1001-10-3	62.00	178.00	<2	11.00	1.67	30.00	10.20	7.07	66.70	62.00	10.00	-20.00	0.00
1001-10-4	91.00	175.00	<2	8.00	1.66	26.00	8.90	7.03	56.40	30.00	8.00	-25.00	50.00
1001-10-5	110.00	162.00	2.80	7.00	1.75	28.00	10.00	7.03	60.50	67.00	7.00	-26.00	90.00
1001-10-6	68.00	167.00	2.60	8.00	1.69	26.00	9.00	6.99	72.30	68.00	8.00	-26.00	98.00
1001-10-7	69.00	175.00	3.10	9.00	1.62	28.00	10.00	6.97	80.70	65.00	9.00	-20.00	0.00
1001-10-8	69.00	175.00	2.70	7.00	1.56	26.00	8.90	6.92	88.70	7.00	9.00	-20.00	0.00
1001-20-1	87.00	178.00	3.60	7.00	1.58	28.00	10.00	6.95	28.40	87.00	9.00	-26.00	90.00
1001-20-2	78.00	>180	<2>	9.00	1.62	<2>	<2>	6.93	52.00	69.00	8.00	-25.00	75.00
1001-20-3	93.00	162.00	<2	8.00	1.68	25.00	10.00	6.99	38.00	66.00	7.00	-26.00	90.00
1001-20-4	77.00	162.00	2.00	8.00	1.62	28.00	10.00	7.01	47.80	<2	7.00	-26.00	<2>
1001-20-5	68.00	178.00	2.30	9.00	1.70	29.00	9.90	6.96	22.40	8.00	8.00	-27.00	0.00
1001-20-6	74.00	>180	<2>	8.00	1.80	<2>	<2>	6.93	38.60	<2	8.00	-26.00	<2>
1001-20-7	79.00	175.00	2.30	8.00	1.76	28.00	10.00	6.96	31.40	8.00	7.00	-25.00	0.00
1001-20-8	82.00	168.00	2.50	10.00	1.80	30.00	10.00	7.16	28.00	20.00	10.00	-19.00	20.00
1021-10-1	69.00	>180	<2>	9.00	1.60	<2>	<2>	7.27	16.70	86.00	8.00	-19.00	77.00
1021-10-2	90.00	166.00	2.90	9.00	1.69	26.00	9.00	7.00	33.50	67.00	8.00	-26.00	92.00
1021-10-3	71.00	168.00	6.00	8.00	1.75	25.00	8.00	6.93	27.60	10.00	6.00	-27.00	8.00
1021-10-4	87.00	177.00	<2	9.00	1.80	29.00	9.00	6.93	36.60	<2	7.00	-26.00	<2>
1021-10-5	78.00	176.00	3.40	12.00	1.61	25.00	8.00	7.07	47.60	8.00	11.00	-19.00	0.00
1021-10-6	91.00	171.00	2.40	9.00	1.58	34.00	9.00	7.07	27.00	69.00	8.00	-22.00	92.00
1021-10-7	80.00	168.00	3.40	10.00	1.67	29.00	10.00	7.06	31.50	66.00	9.00	-23.00	92.00
1021-10-8	79.00	172.00	2.90	10.00	1.61	29.00	9.00	7.06	31.90	10.00	9.00	-23.00	0.00
1021-10-9	96.00	166.00	2.30	9.00	1.59	26.00	9.00	7.07	27.40	69.00	8.00	-22.00	0.00
1021-10-10	73.00	167.00	2.30	9.00	1.60	29.00	9.00	7.07	27.40	10.00	8.00	-22.00	0.00
1021-20-1	79.00	178.00	2.60	7.00	1.70	26.00	9.00	6.93	28.70	22.00	6.00	-26.00	87.00
1021-20-2	77.00	167.00	2.70	8.00	1.70	29.00	10.00	6.93	36.60	9.00	7.00	-25.00	2.00
1021-20-3	76.00	177.00	2.00	8.00	1.66	30.00	10.00	6.89	36.60	69.00	7.00	-27.00	0.00
1021-20-4	91.00	>180	<2>	8.00	1.80	<2>	<2>	6.87	37.10	11.00	7.00	-27.00	2.00
1021-20-5	60.00	172.00	6.00	7.00	1.69	25.00	7.00	6.98	27.00	10.00	6.00	-25.00	10.00
1021-20-6	75.00	168.00	2.90	9.00	2.06	29.00	9.00	7.01	35.00	<2	8.00	-25.00	<2>
1021-20-7	76.00	172.00	2.90	11.00	1.94	30.00	10.00	7.02	37.40	8.00	10.00	-21.00	2.00
1021-20-8	79.00	168.00	2.70	###	1.60	28.00	10.00	###	###	10.00	###	###	###
1021-20-9	72.00	170.00	3.20	10.00	1.76	26.00	9.00	7.03	30.00	<2	8.00	-25.00	<2>
1021-20-10	78.00	167.00	<2	10.00	1.70	28.00	10.00	6.98	37.20	65.00	8.00	-23.00	75.00
1021-10-1	82.00	170.00	2.50	8.00	1.72	27.00	10.00	6.96	40.40	8.00	8.00	-24.00	2.00
1021-10-2	77.00	174.00	2.50	7.00	1.81	30.00	10.00	6.97	36.00	8.00	8.00	-27.00	2.00
1021-10-3	66.00	170.00	2.00	7.00	1.76	31.00	10.00	6.97	32.60	10.00	8.00	-27.00	0.00
1021-10-4	74.00	172.00	<2>	8.00	1.62	27.00	10.00	6.95	38.00	<2>	7.00	-25.00	<2>
1021-10-5	84.00	172.00	3.10	8.00	1.83	34.00	12.00	6.93	32.40	<2>	7.00	-26.00	<2>
1021-10-6	77.00	>180	<2>	8.00	1.80	<2>	<2>	6.97	37.30	<2>	7.00	-27.00	<2>
1021-10-7	80.00	172.00	2.70	9.00	1.65	30.00	10.00	6.96	36.00	10.00	8.00	-24.00	0.00
1021-10-8	72.00	172.00	2.80	8.00	1.69	30.00	10.00	6.96	33.00	7.00	7.00	-25.00	0.00

A. analyte 10. Subsound blood chemistry

5-Chain (Clang (CLR= Carrollgen), type3 (BRC)

Trial ID	Chlorum (mg/dL)	Sodium (mmol/L)	Potassium (mmol/L)	$TC(t)_1$ (mmol/L)	Ionized Calcium (mmol/L)	Glucometer (% PVC)	Hb (g/dL)	pH	$PC(t)_2$ (mmHg)	$TC(t)_2$ (mmHg)	$HC(t)_2$ (mmol/L)	Bicarb (mmol/L)	sO_2 (%)
b22-20-1	00.00	000.00	0.00	0.00	0.00	00.00	00.00	0.00	00.00	00.00	0.00	00.00	000
b22-20-2	00.00	000.00	0.00	0.00	0.00	00.00	00.00	0.00	00.00	<00	0.00	00.00	<00
b22-20-3	00.00	000.00	0.00	0.00	0.00	00.00	00.00	0.00	00.00	00.00	0.00	00.00	000
b22-20-4	00.00	000.00	0.00	0.00	0.00	00.00	00.00	0.00	00.00	<00	0.00	00.00	<00
b22-20-5	00.00	000.00	0.00	0.00	0.00	00.00	00.00	0.00	00.00	00.00	0.00	00.00	000
b22-20-6	00.00	000.00	0.00	0.00	0.00	00.00	00.00	0.00	00.00	<00	0.00	00.00	<00
b22-20-7	00.00	000.00	0.00	0.00	0.00	00.00	00.00	0.00	00.00	00.00	0.00	00.00	000
b22-20-8	000.00	000.00	<0	0.00	0.00	00.00	00.00	0.00	00.00	00.00	0.00	00.00	000

Hb = Sandhead mmp
b2 = Mhanic salmon
1-10-1 and similar numbers = Time class copy depth (m) (Lib number
$TC(t)_1$ = Total carbon dioxide
PCV = Packed cell volume
Hb = Hemoglobin
$PC(t)_2$ = Partial pressure of carbon dioxide
PO_2 = Partial pressure of Oxygen
$HC(t)_2$ = Bicarbonate
Bicarb = Base excess
sO_2 = Saturated Oxygen
<00 = amount can not be calculated
zzzz = not detectable